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生物物理

SEIBUTSU BUTSURI

SUPPLEMENT 1-2

Vol.55

第53回年会予稿集

2015.9.13（日）～ 15（火）

金沢大学　角間キャンパス　自然科学本館

主催　一般社団法人　日本生物物理学会

The Biophysical Society of Japan
一般社団法人　日本生物物理学会　http://www.biophys.jp

生物物理 第55巻 SUPPLEMENT1-1
第53回日本生物物理学会年会 プログラム集
訂正・変更一覧

※2015年9月7日 現在

〔口頭発表〕

頁・番号	訂正・変更内容	訂正・変更前	訂正・変更後
S94 2C1530	共著者追加	○鈴木 康孝1, 守友 博紀1, 藤井 旺成1, 白石 崇人2（1山口大学大学院医学系研究科, 2山口大学理学部） ○Yasutaka Suzuki1, Hiroki Moritomo1, Akinari Fuji1, Takato Shiraishi2 (1Graduate School of Medicine, Yamaguchi University, 2Faculty of Science)	○鈴木 康孝1, 守友 博紀1, 藤井 旺成1, 白石 崇人2, 川俣 純1（1山口大学大学院医学系研究科, 2山口大学理学部） ○Yasutaka Suzuki1, Hiroki Moritomo1, Akinari Fuji1, Takato Shiraishi2, Jun Kawamata 1 (1Graduate School of Medicine, Yamaguchi University, 2Faculty of Science)
S95 2C1545	共著者追加 所属機関変更	○稲垣 成矩1, 松田 知己1, 新井 由之1, 神野 有香2, 筒井 秀和2,3, 岡村 康史2 （1阪大・産研, 2阪大・医学, 3北陸先端大・マテリアル） ○Shigenori Inagaki1, Tomoki Matsuda1, Yoshiyuki Arai1, Yuka Jinno2, Hidekazu Tsutsui2,3, Yasushi Okamura2 （1ISIR., Univ. Osaka, 2Grad. Sch. Med., Univ. Osaka, 3Sch. Mat. Sci., JAIST)	○稲垣 成矩1, 松田 知己2, 新井 由之2, 神野 有香3, 筒井 秀和3,4, 岡村 康史3, 永井 健治1,2 （1阪大・生命機能, 2阪大・産研, 3阪大・医学, 4北陸先端大・マテリアル） ○Shigenori Inagaki1, Tomoki Matsuda2, Yoshiyuki Arai2, Yuka Jinno3, Hidekazu Tsutsui3,4, Yasushi Okamura3, Takeharu Nagai1,2 （1FBS., Univ. Osaka, 2ISIR., Univ. Osaka, 3Grad. Sch. Med., Univ. Osaka, 4Sch. Mat. Sci., JAIST)

〔ポスター発表〕

S103 1Pos015	発表日変更	1Pos015	3Pos231
S126 2Pos127	所属機関スペルミス	1SOKA University Department of Bioinfomatics, 2SOKA University Graduate School of Engeneering	1SOKA University Department of Bioinformatics, 2SOKA University Graduate School of Engineering
S145 3Pos188	発表取り消し	3Pos188	–
S147 3Pos211	発表取り消し	3Pos211	–

年会プログラム集に上記の修正がございます。

第53回年会実行委員会

第 53 回日本生物物理学会年会

The 53rd Annual Meeting of the Biophysical Society of Japan

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The 53rd Annual Meeting of the Biophysical Society of Japan (BSJ2015)

第53回日本生物物理学会年会（2015年度）

会 期 ：
Date：

2015年 9月13日（日） ー 15日（火）
September 13 (Sun) — 15 (Tue), 2015

会 場 ：
Venue：

金沢大学 角間キャンパス 自然科学本館
（石川県金沢市角間町）
Natural Science and Technology Main Hall, Kanazawa University Kakuma Campus
(Kakuma-machi, Kanazawa, Ishikawa 920-1192 Japan)

年会実行委員長：
Chair：

安藤 敏夫
（金沢大学理工研究域数物科学系）
Toshio Ando
(Kanazawa University)

HOMEPAGE　http://www.aeplan.co.jp/jbp2015/

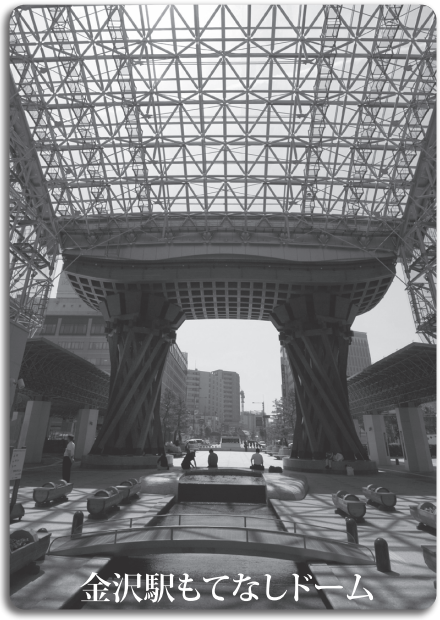
※会期後は日本生物物理学会にデータが移行されますので、学会ホームページよりご覧ください。

抄録本文（Abstract）...

オンライン講演予稿集は、こちらからダウンロードいただけます。

http://www.biophys.jp/dl/pro/53rd_proceedings.pdf
ID：ambsj53　Password ： kanazawa2015

※スマートフォン・タブレット端末向けのプログラム検索・要旨閲覧アプリも現在制作中です。
（2015年 9月7日（月）公開予定）


金沢駅もてなしドーム


金沢城

（写真提供：金沢市）

編集・発行 ： 第53回日本生物物理学会年会実行委員会

第53回日本生物物理学会年会事務局
石川県金沢市角間町
金沢大学理工研究域数物科学系 安藤研究室
E-mail: 53bp-nenkai@ml.kanazawa-u.ac.jp
発行日：表4（裏表紙）記載

The 53rd Annual Meeting of the Biophysical Society of Japan (BSJ2015)

第53回日本生物物理学会年会（2015年度）

開催にあたって

第53回年会 実行委員長
安藤 敏夫
（金沢大学理工研究域数物科学系）

一般社団法人日本生物物理学会第53回年会を2015年9月13 日から15 日の日程で、金沢大学角間キャンパス自然科学本館で開催します。年会では、38 のシンポジウムと約1,000の一般発表が予定されています。金沢で年会を開催するのは学会創立以来、初めてのことです。この時期の金沢は未だ蒸し暑さが残ると予想されますが、暑さを忘れる程に熱気ある議論が行われるものと期待しています。

金沢年会では、例年通り、すべての発表は英語で行います。一般発表では、ポスター発表に加え、久しぶりに口頭発表が復活します。応募者の希望に沿って振り分けましたが、約3対1の割合になりました。シンポジウムには多数のご応募を頂きましたが、限られた枠数のため若手研究者や男女共同参画、海外招待講演者などに配慮して選考させて頂きました。また、生物物理学分野で大型プロジェクトを組織している研究者に積極的にシンポジウムを企画していただきました。生物物理学研究の最近の潮流はもとより、萌芽的研究や勢いを持続している研究など幅広い領域をカバーしています。これらの恒例となっている企画に加え、学生や若手研究者の今後のキャリア構築の一助となるよう、キャリア支援説明会を新たに企画しました。学術研究の議論の場としての年会に新しい機能を持たすことも学会として取り組む課題だと思います。また、年会の国際化に向けた更なる一歩として、未だ十分ではありませんが、プログラム集や年会ホームページ中の英語表記の部分を例年より若干増やしました。

物理的科学の方法一般と生物的科学との融合により生命現象の基本的理解を目指す生物物理学は生命科学の発展に大きく貢献してきました。年会における活発な研究発表、議論が今後の生物物理学の革新、発展の芽となるようにと期待しています。また、最新計測機器などの展示やランチョンセミナーが研究活動の一助になることも期待しています。

なお、金沢にはおいしいお酒や食べ物、伝統工芸、日本庭園、古い街並みなど多くありますので、滞在時間を有効に利用されて、観光の街・金沢を楽しんで頂けたらと思います。

■ 交通のご案内

交通案内

■東京方面から

JR 東京駅 ― 北陸新幹線 約 2 時間 30 分 ― JR 金沢駅 ― 北陸鉄道バス 約35分 ― 金沢大学（自然科学本館）

羽田空港 ― 1時間 ― 小松空港
- 小松バス 約 12 分 ― JR 小松駅 ― 北陸本線 普通 約 30 分 ― JR 金沢駅
- （金沢駅直通）北陸鉄道バス スーパー特急約 40 分 ― JR 金沢駅
- （金沢市内経由金沢駅行き）北陸鉄道バス スーパー特急 約 40 分 ― 香林坊 ― 徒歩5分 ― 香林坊 ①バス停 四校記念館前 ― 北陸鉄道バス 約 20 分 ― 金沢大学（自然科学本館）

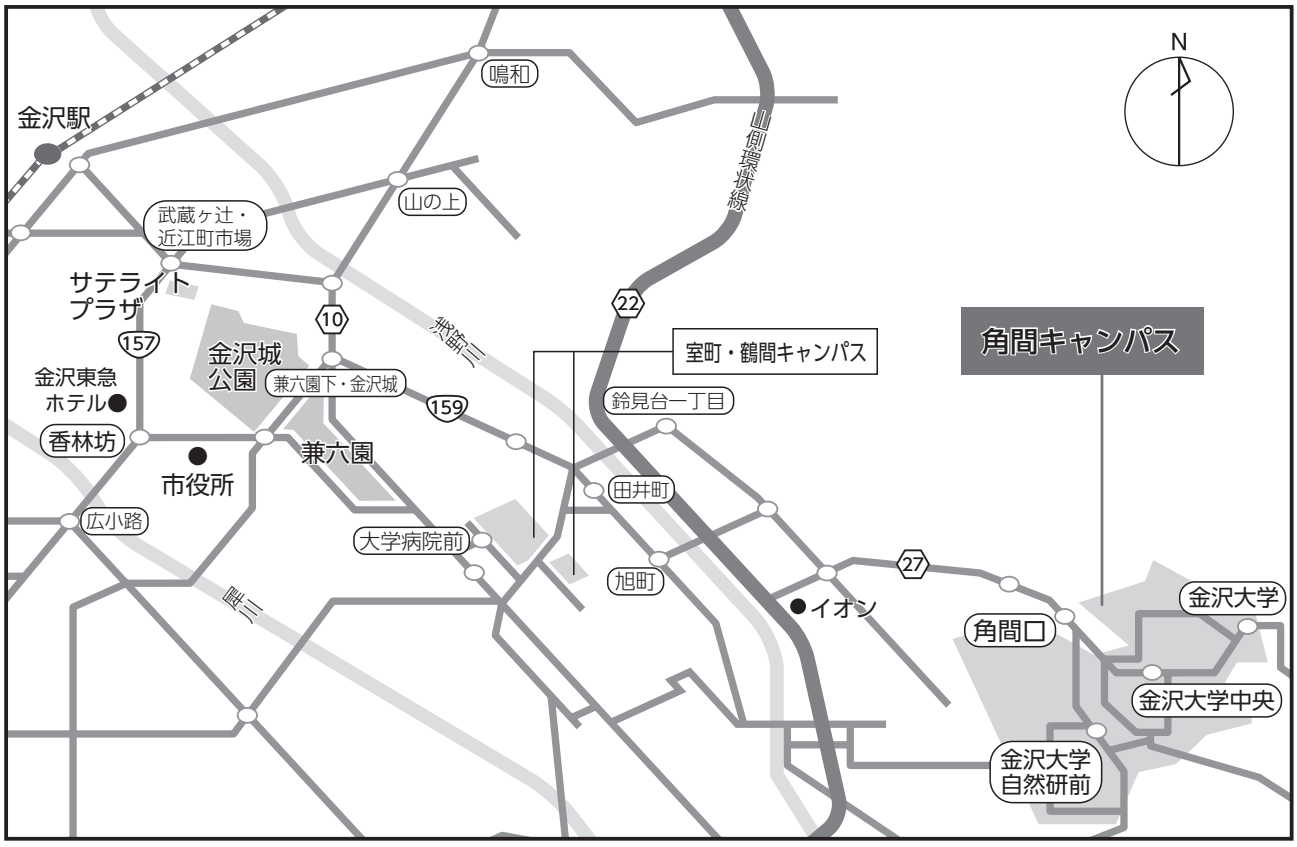
■大阪／京都方面から

JR 大阪駅・京都駅 ― 特急サンダーバード 約 2 時間 30 分 ― JR 金沢駅 ― 北陸鉄道バス 約35分 ― 金沢大学（自然科学本館）

■名古屋方面から

JR 名古屋駅 ― 特急しらさぎ 約 3 時間 ― JR 金沢駅 ― 北陸鉄道バス 約35分 ― 金沢大学（自然科学本館）

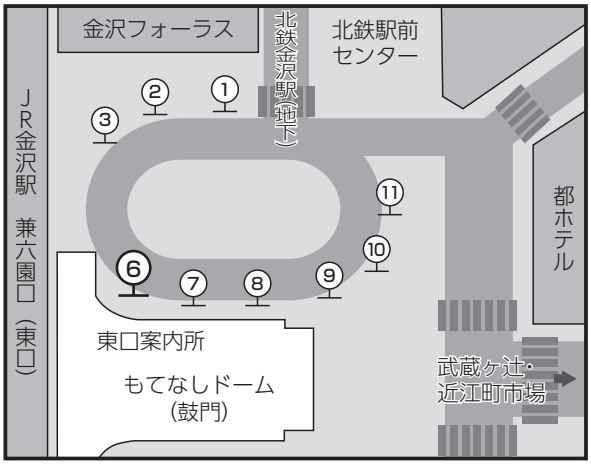
金沢市内案内



金沢大学角間キャンパスの交通案内

実行委員会では、北陸鉄道バスへ依頼をし、年会会期中の「金沢駅」および「金沢大学自然研前」発着の路線バスを以下の通り増便いたしました。

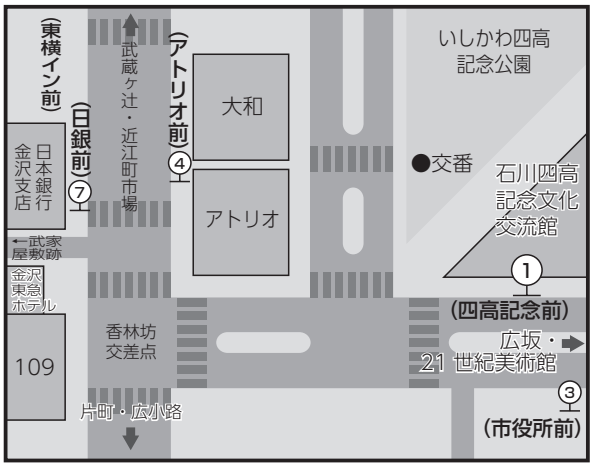
増 便 日：9月13日（日）～15日（火）
増便時間：金沢駅発　7時50分～8時30分
乗 り 場：金沢駅兼六園口（東口）⑥番
増便時間：金沢大学自然研前発　プログラム終了後
乗 り 場：金沢大学自然研前
（台数に制限がございます。ご容赦ください）
※定期便もございますのでご利用ください。
運　　賃：360円



■金沢駅東口より

乗　り　場：金沢駅兼六園口（東口）⑥番
行先・系統：91（平日のみ）
93・94・97　金沢大学行き
降車バス停：金沢大学自然研前
所 要 時 間：約35分
運　　賃：360円

タクシーをご利用の場合　金沢駅からのご利用で金沢大学角間キャンパスまで　約3,500円です。

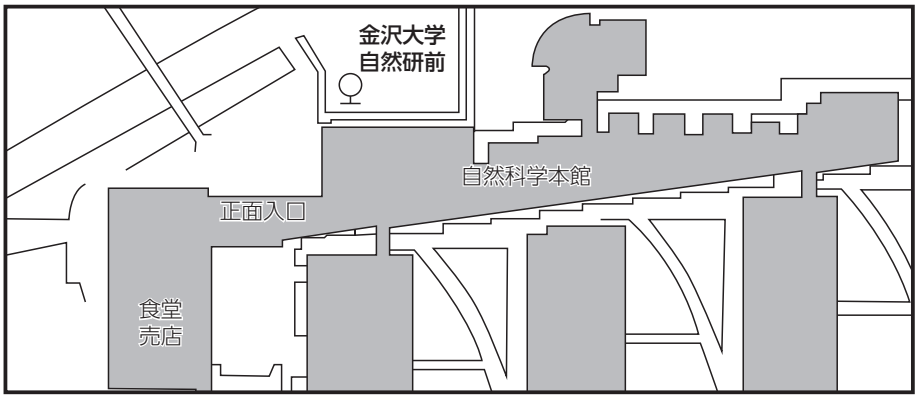


■香林坊より

乗　り　場：香林坊①番（四高記念館前）
行先・系統：93・94・96・97　金沢大学行き
降車バス停：金沢大学自然研前
所 要 時 間：約20分
運　　賃：360円

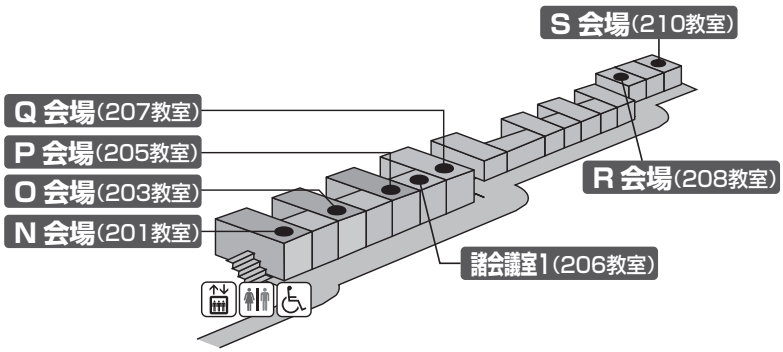
■金沢駅東口まで

プログラム終了後に大学から金沢駅行きバスが増便されます。
※定期便もございますのでご利用ください。
乗　り　場：金沢大学自然研前
所 要 時 間：約35分
運　　賃：360円
※金沢大学内はバスは循環ではありません。終点「金沢大学」で回送となりますので、（自然研前からは）「金沢大学」行のバスにご乗車にならないよう、ご注意ください。

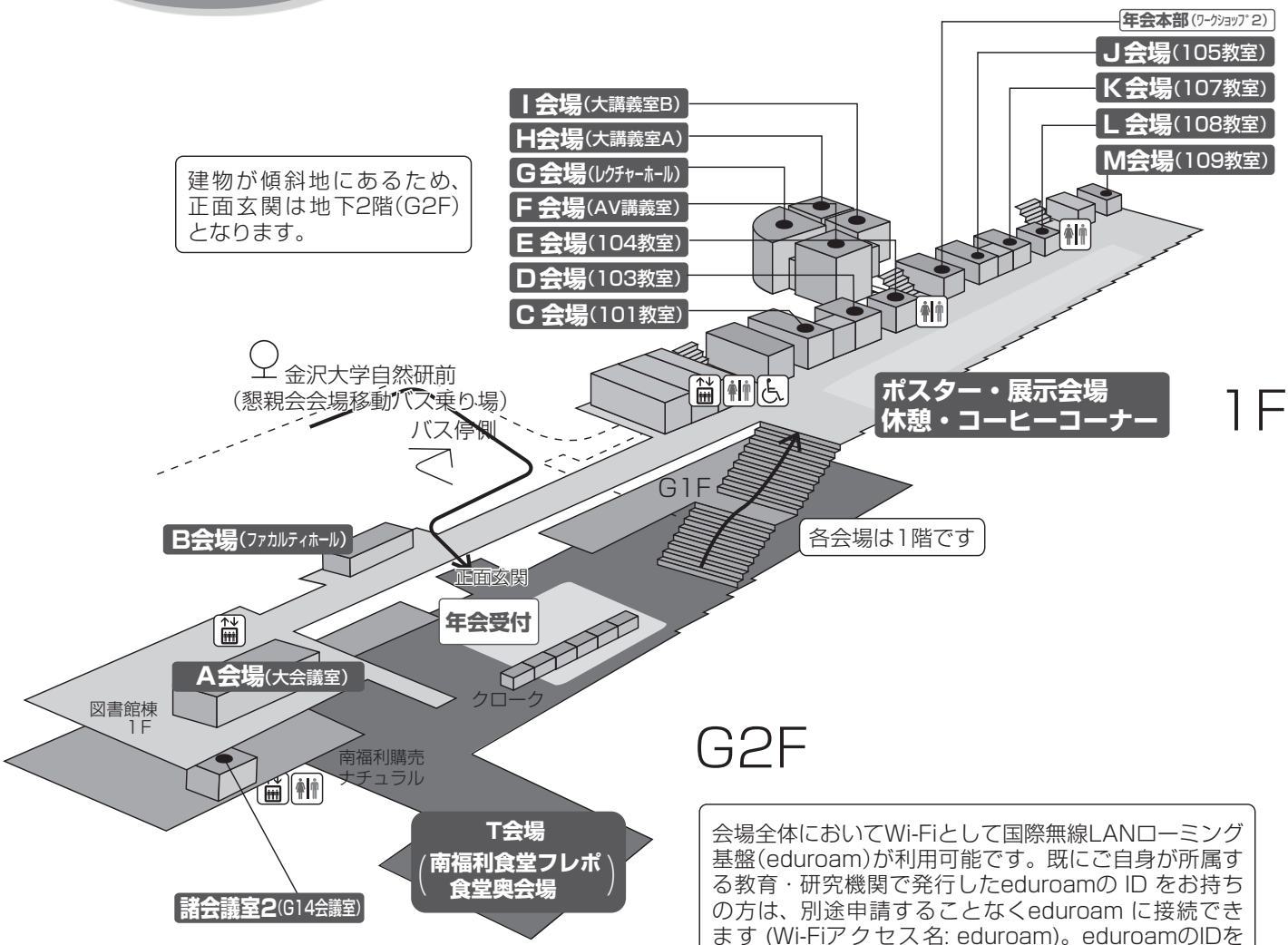


■ 会場のご案内（自然科学本館）

2F

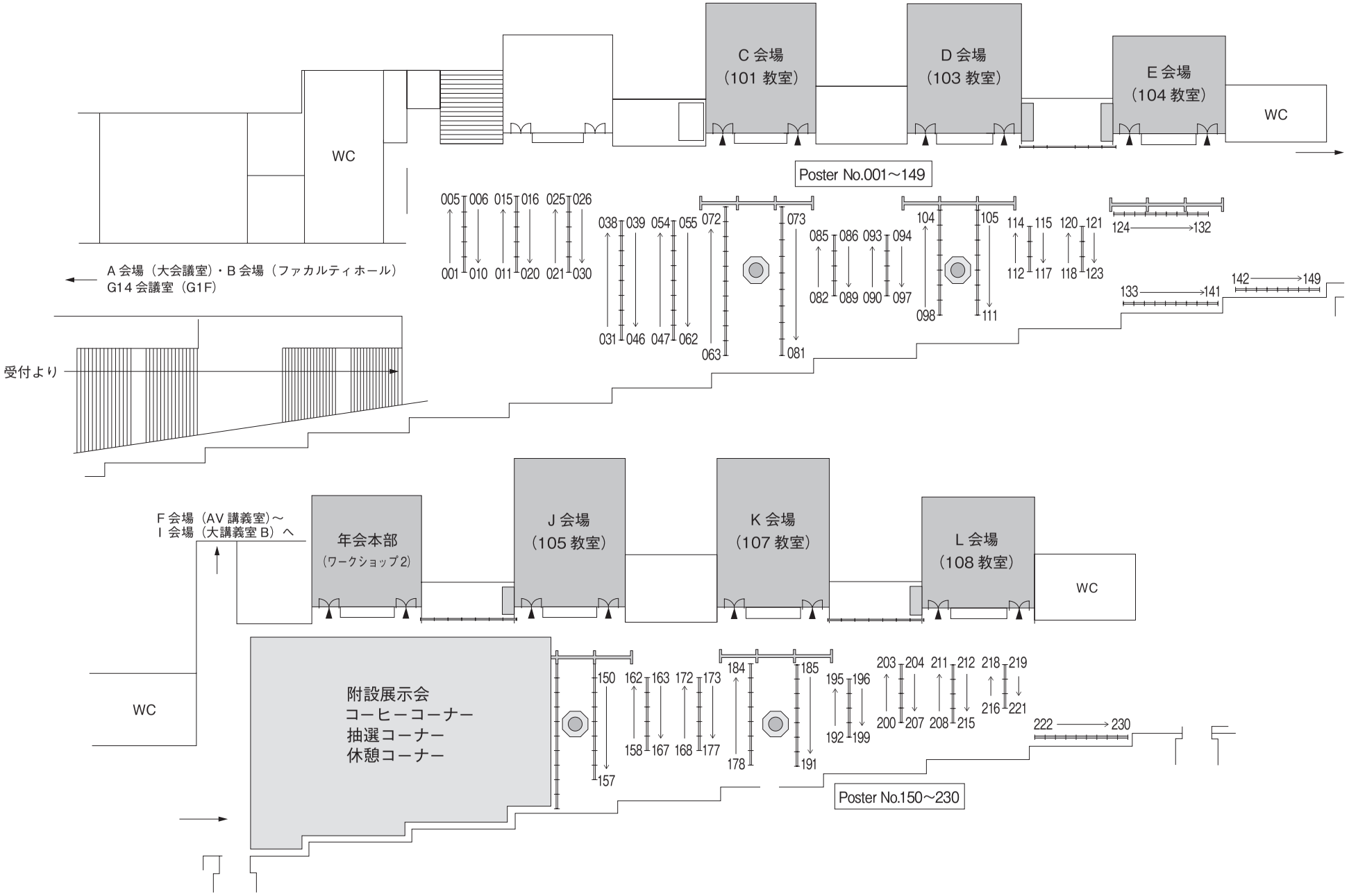


1F/G1F/G2F



会場全体においてWi-Fiとして国際無線LANローミング基盤(eduroam)が利用可能です。既にご自身が所属する教育・研究機関で発行したeduroamの ID をお持ちの方は、別途申請することなくeduroam に接続できます (Wi-Fiアクセス名: eduroam)。eduroamのIDをお持ちでない方は、年会期間のみ有効なeduroam-IDと接続パスワードを年会受付で配布します。
※場所によっては、電波が微弱なところもあります。
※講演中のインターネットご利用はご遠慮ください。

■ ポスター会場のご案内（1F）



2015年9月13日（日）：年会1日目

English

金沢大学 角間キャンパス 自然科学本館

			9：00	10：00	11：00	12：00	13：00	14：00	15：00	16：00	17：00	18：00	19：00
1F	大会議室	A会場	1SAA べん毛・繊毛が織りなす多様な生命現象に挑む ～分子から個体まで～					1SAP 温度生物学の幕開け					
1F	ファカルティホール	B会場	1SBA アルキンタグ等ラマンプローブをはじめとする 小分子バイオイメージング最前線			11：50	12：40	1SBP 蛋白質・核酸共存系に対する実験家と理論家の挑戦					
1F	101教室	C会場	1SCA 生体分子活性サイトの新しい3D 構造決定法と機能解析			キャリア支援合同説明会 （ランチョン形式） 11：50－12：40		1C バイオイメージング I					
1F	103教室	D会場	1SDA 生命科学における大規模ネットワーク解析					1D 光生物：視覚・光受容 I					
1F	104教室	E会場				Biophysics and Physicobiology 論文賞受賞講演 12：50－13：20		1E 蛋白質：構造・構造機能相関 II		16：15			
1F	AV 講義室	F会場	若手招待講演			科研費説明会 （ランチョン形式）		1SFP 膜タンパク質と膜脂質が織りなす協奏と合奏					
1F	レクチャーホール	G会場	1SGA 次世代スパコン「ポスト京」が拓く バイオスーパーコンピューティング			1LG 浜松ホトニクス 株式会社		1SGP X 線自由電子レーザー施設 SACLA が拓く生物物理研究の新展開					
1F	大講義室 A	H会場	1SHA 生命現象の基本に迫る 動的クロマチン構造・機能研究の最前線			1LH 株式会社菱化 システム		1SHP チューブリンの構造スイッチングとその生理的意義					
1F	大講義室 B	I会場	1SIA 自然史に学ぶバイオミメティクスの未来			1LI 日本蛋白質構造 データバンク		1SIP 超解像光学顕微鏡のライフサイエンスへの展開			16：20		
1F	105教室	J会場	1SJA アクチン線維と結合タンパク質の構造と機能とゆらぎ			11：50	12：40	1J 細胞生物的課題 I					
1F	107教室	K会場	1SKA 膜を介したプロトン透過機構					1K 分子モーター I					
1F	108教室	L会場	9：00		11：30			1L 蛋白質：機能・計測・解析の方法論					
1F	109教室	M会場						1M 蛋白質：構造・構造機能相関 I					
2F	201教室	N会場						1Na バイオエンジニアリング 13：20－14：20	1Nb 膜蛋白質 I 14：25－16：15				
2F	203教室	O会場						1O 核酸結合蛋白質					
2F	207教室	Q会場						1Qa 生体膜・人工膜 I 13：20－15：25	1Qb 非平衡・生体リズム 15：30－16：15				
2F	210教室	S会場						キャリア支援個別説明会 13：20－14：32					
2F	206教室	諸会議室1				Biophysics and Physicobiology 編集委員会 11：40－12：40	13：20		若手賞選考委員会 15：00－16：00				
G1F	G14会議室	諸会議室2			科研費説明会 打ち合わせ 11：00－11：45								平成27年第2回理事会 18：30－19：30
1F	プロムナード	ポスター会場	ポスター貼付・掲示 8：45－16：30							ポスター討論 奇数 16：30－17：30	ポスター討論 偶数 17：30－18：30	撤去	
1F	プロムナード	企業展示	機器・試薬・書籍展示 9：30－18：30										

2015年9月14日（月）：年会2日目

English

金沢大学　角間キャンパス　自然科学本館

			9：00	10：00	11：00	12：00	13：00	14：00	15：00	16：00	17：00	18：00	19：00
1F	大会議室	A会場	2SAA 動的構造生命科学を拓く新発想測定技術			2LA 日本エフイー・アイ株式会社		2SAP 生体マシナリーにおける力発生と進化の共通原理					
1F	ファカルティホール	B会場						2B 蛋白質：構造・構造機能相関 IV 13：55−16：00					
1F	101教室	C会場	2Ca 分子モーターII 8：45−10：00					2Cb バイオイメージング II		16：15			
1F	103教室	D会場	2SDA 脂質活性構造研究の最前線					2D 光生物：視覚・光受容 II					
1F	104教室	E会場				会員総会ワークショップ 12：20−13：40		2E 蛋白質：物性					
1F	AV 講義室	F会場	2SFA 大自由度ダイナミクスから“生きている状態”の記述へ			2LF オリンパス株式会社	キャリア支援合同説明セミナー 12：30−13：40	2SFP ナノとマクロを繋ぐ生物電子顕微鏡アプローチ					
1F	レクチャーホール	G会場	2SGA 生体分子の機能を制御する柔らかさ			2LG オックスフォード・インストゥルメンツ株式会社		2SGP 蛋白質の相互作用と動的立体構造変化					
1F	大講義室 A	H会場	2SHA 新生鎖の合成と構造形成過程に潜む生物物理学			分野別専門委員会 出版倫理講演会		2SHP ポンプ、酵素、モーター、機能の鍵：pKa					
1F	大講義室 B	I会場	2SIA ATP 加水分解が介在する時空間動秩序の形成			11：30	12：20	2SIP 高次の生命現象を可能にする類似構造による多様なリガンド認識機構		16：25			
1F	105教室	J会場	2SJA 物質と生命の境界を探る合成生物学					2Ja 細胞生物的課題 II 13：55−15：10	2Jb ヘム蛋白質 15：15−16：15				
1F	107教室	K会場	2SKA GPCR の多様性、そしてその機能理解へ至る多様なアプローチ					2K 蛋白質：構造・構造機能相関 III 13：55−16：15					
1F	108教室	L会場	8：45		11：15			2L 生命情報科学 13：55−15：10					
1F	109教室	M会場						2Ma 筋肉	2Mb 神経科学 15：15−16：00				
2F	201教室	N会場						2Na 膜蛋白質 II	2Nb 発生・分化				
2F	203教室	O会場						2Oa 光生物：光合成	2Ob 核酸：構造・物性 15：15−16：15				
2F	205教室	P会場						2P 数理生物学					
2F	207教室	Q会場					13：55	2Q 生体膜・人工膜 II					
2F	210教室	S会場						キャリア支援個別説明会 14：00−16：00					
2F	206教室	諸会議室1		企業との意見交換会 9：30−11：00						若手の会会議 16：00−17：00			
1F	南福利食堂フレポ　奥	T会場			男女共同参画・若手支援委員会 10：00−11：30	男女共同参画・若手支援シンポジウム 11：30−12：20							
1F	プロムナード	ポスター会場	ポスター貼付・掲示 8：30−16：30								ポスター討論 奇数 16：30−17：30	ポスター討論 偶数 17：30−18：30	撤去
1F	プロムナード	企業展示		機器・試薬・書籍展示 9：30−18：30									

懇親会（金沢東急ホテル） 19：30−21：30

2015年9月15日（火）：年会3日目

English

金沢大学 角間キャンパス 自然科学本館

1F

		9：00	10：00	11：00	12：00	13：00	14：00	15：00	16：00	17：00	18：00	19：00
大会議室	A会場	3SAA 細胞に近づく構造生命科学の最前線										
ファカルティホール	B会場	3SBA 最新ハイブリッドアプローチによる機能構造解析										
101教室	C会場	3SCA 電気生理学的アプローチによる膜タンパク質構造機能相関										
103教室	D会場	3SDA 輸送膜タンパク質のダイナミクス										
104教室	E会場	9：00			12：00							
AV講義室	F会場	3SFA 細胞を診て操作する生物物理的アプローチ				3LF 株式会社 オプトライン						
レクチャーホール	G会場	3SGA 理論と実験の統合的アプローチが解き明かす生体秩序構造 ー分子から組織までー				3LG 株式会社生体分子計測研究所						
大講義室A	H会場	3SHA 少数分子が担う生命現象				3LH 株式会社 ニコンインステック						
大講義室B	I会場	3SIA タンパク質を活かす多成分システムの分子レベル解析：水和効果から細胞内環境へ			12：15	13：05						
105教室	J会場	3SJA 生体分子 におけるレアイベントの探求										
107教室	K会場	3SKA 人工細胞を創る・動かす・活用する										
プロムナード	ポスター会場	ポスター貼付・掲示 8：45ー13：30					ポスター討論 奇数 13：30ー14：30	ポスター討論 偶数 14：30ー15：30	撤去			
	企業展示	機器・試薬・書籍展示 9：30ー15：45										

参加者へのご案内

1. 年会受付と参加登録

◇ 年会受付

場　　所: 自然科学本館 G2 階入口アカデミックホール(「会場のご案内」6 ページを参照)
受付時間: 9 月 13 日(日) 8:30 – 17:00
14 日(月) 8:15 – 17:00
15 日(火) 8:30 – 15:00

◆事前登録

事前登録が完了された方は、日本生物物理学会会員・非会員共に参加証および領収証、プログラム集冊子が事前送付されますので、会場での受付は不要です。当日は必ず参加証をお持ちください。
但し、海外からの参加者は、年会受付にて参加証とプログラム集をお受け取りください。

※ネームタグホルダーを当日配布しますので、会場内では必ず参加証をご着用ください。
配付場所: 年会受付付近、一部の講演会場前

注意 1) 事前登録は年会参加登録費(参加費)の振込後に完了します。振込がない場合、オンライン登録は無効となります。当日受付で当日参加費をお支払いください。
注意 2) 日本生物物理学会会員は年度会費を納めていない場合、参加証が送付されません。年度会費未納者・新規入会受付デスクにて年度会費をお支払いください。
注意 3) 参加費・年度会費ともに振込済みで、参加証が事前送付されていない場合は、総合受付デスクまでお越しください。
注意 4) 非会員のシンポジウム招待講演者については、会員である必要はなく、また、登録費は免除されます。懇親会に無料でご招待します。
注意 5) 海外の機関に所属する非会員については、ご入会いただかずに一般発表をしていただけます。

◆当日登録

事前登録が完了していない方は当日登録をしていただきます。
当日受付にお越しの上、参加費を現金でお支払いください。

◇ 当日年会諸費用 (一覧表)

当日参加	会員				非会員		
	正会員	シニア会員	大学院生	学部学生	一般	大学院生	学部学生
当日参加費 Registration	¥9,000	¥5,000	¥5,000	¥0	¥12,000	¥6,000	¥0
懇親会費 Banquet	¥7,000	¥5,000	¥5,000	¥3,000	¥7,000	¥5,000	¥3,000

・参加のみの学部学生は参加費無料です。当日受付で学生証を提示してください。
参加証とプログラム集冊子をお渡しします。ただし、懇親会は有料です。

・若手招待講演者、Biophysics and Physicobiology 論文賞受賞講演者、Biophysics and Physicobiology Editors' Choice Award 受賞代表者は、懇親会に無料でご招待します。
既に懇親会参加費を振り込まれている場合は、総合受付デスクで返金します。

◇ 参加証（名札）

参加証は会場内では必ずご着用ください。参加証のない方のご入場は固くお断りいたします。
事前送付された参加証は必ず会場にお持ちください（ネームタグホルダーは会場内で配布いたします）。

◇ 領収書の発行

参加証とともに領収書をお渡しいたしますが、別の形式の領収書が必要な場合、お渡しした領収書と引き換えに総合受付デスクにて発行いたします。

◇ プログラム集冊子/オンライン予稿集【8 月 18 日（火）公開予定】

プログラム集冊子（前付・プログラム）は日本生物物理学会会員・事前登録が完了された非会員に事前に送付いたします（プログラム集冊子は総合受付デスクでも当日販売（3,500 円/税込）を行います）。なお予稿本文はプログラム集冊子には掲載されません。予稿本文は、オンライン予稿集をダウンロードして閲覧いただくことになります。

オンライン予稿集：
http://www.biophys.jp/dl/pro/53rd_proceedings.pdf
ダウンロード ID:ambsj53
パスワード：kanazawa2015

プログラム（タイトル、発表者、所属）と予稿集は、年会ホームページにて公開します。
年会終了後は、半年ほど経て日本生物物理学会ホームページの年会の記録にて予稿集の pdf ファイルが公開される他、CiNii（国立情報学研究所の論文情報ナビゲータ）にリンクが張られ、
CiNii の生物物理のページでも予稿本文が公開されます。
日本生物物理学会ホームページの年会の記録(http://www.biophys.jp/ann/ann02.html)
CiNii の生物物理のページ(http://ci.nii.ac.jp/organ/journal/INT1000001547_jp.html)

◇ プログラム検索（ウェブ版）【8 月 18 日（火）公開予定】

年会ホームページより「プログラム検索」を公開します。項目［演題タイトル（和文・英文）、発表者名（共著者含む）（漢字、カナ、ローマ字）、発表形式］から、演題番号、発表日、会場を検索・表示します。

◇ プログラム検索・予稿閲覧アプリ（無料）【9 月 7 日（月）公開予定】

スマートフォン(iPhone/Android)やタブレット(iPad/iPod Touch/Android)端末に対応した予稿閲覧アプリをご利用いただけます（演題検索、タイムテーブル一覧表示、ブックマーク登録等）。
App Store、Google Play よりダウンロードしてください（無料）。年会ホームページにもアプリ提供サイト(App Store, Google Play)を掲載しております。

アプリケーション名：第 53 回日本生物物理学会年会
検索ワード：BSJ2015、生物物理、日本生物物理学会
アプリケーションの予稿閲覧パスワード：kanazawa2015

◇ 年度会費の支払いと入会の手続き

日本生物物理学会の年度会費が未納の場合は、年会受付の年度会費未納者・新規入会受付デスクでお支払いください。また、日本生物物理学会への新規入会も受け付けます。

2. 会場内のサービス・施設

◇ クローク

場　　所: 自然科学本館 G2 階入口アカデミックホール(「会場のご案内」6 ページを参照)

利用時間: 9 月 13 日(日) 8:30 – 19:00
14 日(月) 8:15 – 18:50
15 日(火) 8:30 - 16:00

※貴重品や傘、またコンピュータなどについては、破損、紛失などの責任は負いかねますので、各自でお持ちください。
※懇親会への移動など会場を去られる際は荷物をお引き取りください。

◇ 昼食

ランチョンセミナー(1～3 日目)、科研費説明会(1 日目)、男女共同参画・若手支援シンポジウム(2 日目)でお弁当とお茶が無料で提供されます。当日の午前中に整理券を配布いたします。整理券のご利用方法は次ページ「ランチョンセミナー」をご参照ください。
この他、分野別専門委員会(2 日目 英文誌「Biophysics and Physicobiology」セミナー、次ページ参照)を開催し、お弁当とお茶が無料で提供されます(整理券なし・数量に限りがあります)。積極的にご参加ください。
また会期中以下の食堂と購買店舗をご利用いただけます。

■南福利食堂 フレポ(G2 階)
11:30 – 14:00(注文 13:30 まで、3 日間とも)
※17:30 - 20:00 の時間帯にも営業します(但し、14 日と 15 日のみ)
■南福利食堂 すみれ亭(自然科学系図書館棟 2 階)
11:00 - 14:30(注文 14:00 まで、3 日間とも)
■南福利購買店舗 ナチュラル(フレポの向かい)
9:00 - 17:00 (3 日間とも)

◇ 呼び出し

会場内での呼び出しは、緊急の場合を除いて一切行いません。参加者間の連絡用として、年会受付付近に伝言板を設置しますので、ご利用ください。

◇ 駐車場

会場には参加者用駐車場はございません。会場へは公共交通機関をご利用ください。

◇ 宿泊

宿泊に関しては年会ホームページ「宿泊案内」をご参照ください。

◇ インターネット

会場全体において Wi-Fi として国際無線 LAN ローミング基盤(eduroam) がご利用いただけます。既にご自身が所属する教育・研究機関で発行した eduroam の ID をお持ちの方は、別途申請することなく eduroam に接続することが可能です (Wi-Fi アクセス名: eduroam)。eduroam の ID をお持ちでない方は、年会期間のみ有効の eduroam-ID と接続パスワードを年会受付で配布します。

※建物の構造や電波状況によっては接続できない場合もあります。eduroam-ID は 1 人につき 1 つのみ発行いたします。

◇ コーヒーコーナー

ポスター・展示会場休憩スペース(1 階 プロムナード)をご利用ください。

◇ 託児所

年会期間中は、託児所を設置いたします。詳しくは年会ホームページをご覧ください。

3. 年会行事・プログラム

◇ 会員総会・第 2 回ワークショップ

一般社団法人日本生物物理学会第 2 回会員総会を年会 2 日目、9 月 14 日(月)12:20-13:40 に H 会場(大講義室 A)で開催しますのでご出席ください。また、第 2 回会員総会ワークショップを開催します。詳しくは 7. 開催通知(21 ページ)をご覧ください。

◇ 若手招待講演

日本生物物理学会若手奨励賞の選考会である講演会(若手招待講演)を、年会 1 日目 9 月 13 日(日) 9:00 – 11:30 に F 会場(AV 講義室)で開催します。

◇ Biophysics and Physicobiology 論文賞受賞講演

Biophysics and Physicobiology 論文賞受賞の講演会を、年会 1 日目 9 月 13 日(日)12:50 - 13:20 に H 会場(大講義室 A)で開催します。

◇ 懇親会

日時: 9 月 14 日(月)19:30 – 21:30 (年会会場から貸切バスで移動, 発車時刻の予定 18:45 – 19:00)
会場: 金沢東急ホテル 5 階『ボールルーム』(バス停「香林坊」下車 徒歩1分)
(石川県金沢市香林坊 2-1-1) Tel: 076-231-2411
※懇親会の当日参加も受け付けいたします(受付場所: 総合受付デスク、または懇親会会場前)。

◇ 男女共同参画・若手支援シンポジウム

日時: 9 月 14 日(月)11:30 – 12:20
会場: T 会場(自然科学本館 南福利食堂 フレポ(G2 階) 奥)
昼食: お弁当とお茶が無料で提供されます(整理券を配布いたします。次ページを参照)。

◇ 分野別専門委員会(英文誌「Biophysics and Physicobiology」セミナー)

日時: 9 月 14 日(月)11:30 – 12:20
会場: H 会場(大講義室 A)
対象: 分野別専門委員(委員以外の会員の方の参加も歓迎します)
内容: 英文誌「Biophysics and Physicobiology」セミナーとして、高石 雅人 氏(エルゼビア・ジャパン株式会社)による「出版倫理」についての講演があります。
昼食: 委員以外の会員には、先着 20 名までお弁当とお茶が無料で提供されます(整理券なし)。

◇ キャリア支援説明会

日時: 9 月 13 日(日)10:00 - 11:40、13:20 - 16:20(詳細は当日に掲示)
14 日(月) 9:00 - 11:20、14:00 - 16:00 (詳細は当日に掲示)
会場: S 会場(210 教室)
対象: 就職を考えておられる学生や研究者など
内容: 企業、研究機関、研究室の説明、意見交換

◇ 科研費説明会

日時: 9 月 13 日(日)11:50 - 12:40
会場: F 会場(AV 講義室)
昼食: お弁当とお茶が無料で提供されます(整理券を配布いたします。下記参照)。

◇ ランチョンセミナー

昼食(お弁当とお茶、無料)をとりながらの協力企業によるセミナーにご参加ください。なお、お弁当の数に限りがあるため当日の以下の時間帯に整理券を配布いたします。セミナー開始前に、会場入り口で整理券と引き換えにお弁当を受け取り、ご入場ください(整理券の発券方法は下記参照)。

◆整理券の発券について

ランチョンセミナー整理券は整理券配布デスクにて配布いたします。
時間: 9月13日(日)・15日(火) 8:30 – 10:30、14日(月) 8:15 – 10:30
場所: 自然科学本館G2階入口アカデミックホール 年会受付付近
※整理券はランチョンセミナー共催の企業、団体よりご提供いただく昼食の引換券になります。
当日開催されるセミナー分のみ発券いたします。券は枚数が無くなり次第終了となります。

◆整理券の注意事項

整理券は各日、セミナー開始後、無効となります。
午前のプログラム終了後、ランチョンセミナー開始時間までにご来場ください。
セミナー開始までにご来場されない場合、整理券は無効となり、お弁当は整理券をお持ちでない参加者にご提供されますことをご了承ください。

◇ 機器・試薬・書籍等附設展示会

機器、試薬、ソフトウエア、書籍などの附設展示会をポスター・展示会場(1階 プロムナード)で行います。

◇ 市民講演会

テーマ: 生物発光が拓く未来社会
日 時: 9 月 12 日(土) 開場 13:30、 開演 14:00、 終演 16:00
会 場: 石川県教育会館ホール(金沢市香林坊 1-2-40)
JR 金沢駅よりバス 15 分(香林坊下車/徒歩 2 分) JR 金沢駅よりタクシー12 分
参加費: 無料(ポスター「一家に 1 枚 動く!タンパク質」(文科省制作・日本生物物理学会提案)提供)
お問い合わせ: 実行委員会サポート
E-mail: jbp2015@aeplan.co.jp

4. 禁止事項

◇ 撮影・録音

会場内でのカメラ、ビデオ、携帯電話などによる撮影や講演音声の録音などを禁止します。

◇ 喫煙・飲食

会場での喫煙は、所定の喫煙所（自然系図書館 G2階 生協売店北側の外、自然科学1号館 1階 自動販売機コーナー外のテラス）以外では禁止です。講演会場内での飲食はランチョンセミナー、男女共同参画・若手支援シンポジウム、科研費説明会、各種委員会など食事が提供される場合を除いて禁止します。

◇ 携帯電話

シンポジウム、一般口頭発表やポスター発表等の会場内での携帯電話による通話を禁止します。講演会場内では電源をオフにするかマナーモードに設定し、呼び出し音が鳴らないようご注意ください。

5. 年会についての問い合わせ

◇ **会期中**　年会本部 （会期中のみ通じます） Tel: 070-5453-8365

◇ **会期外**　年会実行委員会 E-mail:53bp-nenkai@ml.kanazawa-u.ac.jp

参加登録・演題登録 システムサポートデスク

〒113-0033 東京都文京区本郷2-26-11 浜田ビル5F
中西印刷株式会社 東京営業部内
E-mail: bsj2015sys-sprt@e-naf.jp

年会実行委員会サポート・展示・広告

〒101-0003 東京都千代田区一ツ橋 2-4-4 岩波書店一ツ橋別館 4F
株式会社エー・イー企画 Tel: 03-3230-2744（代表） Fax: 03-3230-2479
実行委員会サポート E-mail: jbp2015@aeplan.co.jp
広 告 ・展 示 関 連 E-mail: e_jbp53@aeplan.co.jp

6. 発表者へのご案内

◇ 使用言語

すべての発表言語は原則として英語をお使いください。

◇ 映写機器

会場にはパソコンを用意いたしません。ご自身のノートパソコンを必ずお持ちください。
発表に使用できる映写機器は、液晶プロジェクターのみです。音声出力には対応しません。
会場に備え付けの液晶プロジェクターにより、図等をスクリーンに映写して発表します。
使用ソフトはパワーポイント（米国マイクロソフト社）を標準とします。画像解像度は 1024×768 ピクセル（XGA）です。この環境下で発表データを作成ください。これより大きい画面サイズでデータを作成すると、スクリーン映写時に画面をはみ出す等の不具合が起こる可能性がある旨ご理解ください。

注意 1）会場スタッフがパソコンを会場に備え付けられた切り替え装置（セレクター）に接続いたします。
注意 2）切り替え装置に繋がるパソコンの映像出力端子は、
「ミニ D-sub15 ピン端子（メス）」のみです。
端子の形状が異なる場合（Macintosh 等）、変換アダプターをお持ちください。
注意 3）発表に使用するパワーポイントファイルが入った USB メモリーを念のためにお持ちください。

注意 4)バッテリー切れに備え、必ず電源アダプターをお持ちください。
注意 5)発表中にスクリーンセーバーや省電力モードにならないよう、設定してください。

◇ シンポジウム、若手招待講演のオーガナイザーの方へ

受付: セッション開始の 15 分前までに各会場の「進行席」までお越しの上、係りの者に来場された旨をお伝えください。

進行: 一任いたしますので、講演者の講演時間を厳守し、円滑な運営にご協力ください。プログラムに記載されている各講演者の講演時間等に変更が生じた場合は、会場内の係りの者にご指示ください。
会場には時間を計測するスタッフを置いています。

◇ シンポジウム、若手招待講演の講演者の方へ

受付: セッション開始の 15 分前までに各会場の「PC 受付」にお越しください。
発表スライドをご確認いただいた後、会場スタッフがパソコンを切り替え装置(セレクター)に接続いたします。※スライドチェック用の試写室は設けておりません。

講演時間: シンポジウムの時間配分はオーガナイザーに一任しております。
若手招待講演の講演時間は、発表 10 分、討論 3 分、パソコンの交換に 2 分です。

若手招待講演の発表プログラムが終了後、発表会場内で若手招待講演証書が授与されます。

◇ 一般口頭発表の座長の方へ

受付: 担当セッションの開始 10 分前までに各会場の「進行席」までお越しの上、係りの者に来場された旨をお伝えください。

進行: 多くの講演者の発表を滞りなく進めるために、時間厳守をお願いします。
会場には時間を計測するスタッフを置いています。

◇ 一般口頭発表の講演者の方へ

受付: ご自身の発表の 20 分前までに指定された会場の「PC 受付」までお越しください。会場スタッフがパソコンを切り替え装置(セレクター)に接続いたします。※スライドチェック用の試写室は設けておりません。

講演時間: 発表 10 分、質疑応答4分、交代時間1分です。

◇ ポスター発表の方へ

ポスターの貼付、展示、説明・討論、撤去:

		9 月 13 日(日)	9 月 14 日(月)	9 月 15 日(火)
貼付・展示		8:45 – 16:30	8:30 – 16:30	8:45 – 13:30
説明・討論	奇数番号	16:30 – 17:30	16:30 – 17:30	13:30 – 14:30
	偶数番号	17:30 – 18:30	17:30 – 18:30	14:30 – 15:30
撤去		18:45 までに撤去	18:45 までに撤去	15:45 までに撤去

1. ポスターは日替わりで貼り替えてください。
2. ポスターボードの大きさは、幅 90 cm、高さ 210 cm。貼付に必要な押しピンは会場に用意します。
3. 撤去時間を過ぎて残ったポスターは年会事務局にて破棄しますので、ご了承ください。

◇ ポスター発表要項

ポスターは英語で作成してください。
ただし、タイトル、所属、著者名は、可能であれば日本語の併記もお願いいたします。
発表代表者の氏名には左肩に小さな○印を付けてください。

◇ 発表形式と演題番号（各予稿左上の番号）の見方

発表形式は、シンポジウム発表（Symposium Speech）、若手招待講演（“Early Career Award in Biophysics” Candidate Presentations）、一般口頭発表（Oral Presentation）、ポスター発表（Poster Presentation）があります。

シンポジウム発表：（例）1SAA-03
1 文字目は発表日（1:9 月 13 日(日)、2:9 月 14 日（月）、3:9 月 15 日（火））、2 文字目は Symposium（S）、3 文字目は会場名（A会場）、4 文字目は午前・午後（AM,PM）、最後の 2 桁の数字は発表順です。

若手招待講演：（例）1YF1045
1 文字目は発表日（1:9 月 13 日(日)、2:9 月 14 日（月）、3:9 月 15 日（火））、2 文字目は Young（Y）、3 文字目は会場名（F 会場）、最後の 4 桁の数字は講演開始時刻です。

一般口頭発表(例) 1D1320
1 文字目は発表日（1:9 月 13 日(日)、2:9 月 14 日（月）、3:9 月 15 日（火））、2 文字目は会場名（D 会場）、最後の 4 桁の数字は講演開始時刻です。

ポスター発表：（例 1）1Pos001
1 文字目は発表日（1:9 月 13 日(日)、2:9 月 14 日（月）、3:9 月 15 日（火））、2 文字目は Poster（Pos）、最後の 3 桁の数字はパネル番号を示します。

7. 一般社団法人日本生物物理学会第 2 回会員総会開催通知

日時: 9 月 14 日（月）12:20 – 13:40
会場: H 会場（大講義室 A）

一般社団法人日本生物物理学会第 2 回会員総会を開催いたします。主な議題は下記の通りです。
是非ご出席ください。

議長: 会長 中村 春木

総会議題

(1) 報告事項

平成 26 年度第二期決算報告ならびに監査結果報告

平成 27 年度会計ならびに事業の中間報告と今後の計画

次期年会について

名誉会員について

(2) 第 2 回会員総会ワークショップ：生物物理が解明する生命のメカニズム

概要：次ページ掲載

第 2 回会員総会ワークショップ：生物物理が解明する生命のメカニズム
(Biophysics reveals the machinery of life)

日時：9月14日(月)12:20 - 13:40　会員総会中
会場：H会場(大講義室A)
形式：講演会

概要 [Abstract]
生物物理学の主題の一つは、生命のメカニズムを解析・操作する「道具」の開発とその「応用」である。今回は、生命の重要なメカニズムの一つである「光合成」に焦点を当てて、自由電子レーザと量子化学計算という2つの「道具」を活用し、国際競争に打ち勝って解明がなされた研究を発表していただき、今後の生物物理学への道しるべとしたい。

One of the major topics in biophysics is to develop and apply tools for understanding the machinery of life. In this workshop, we focus on oxygenic photosynthesis, and the recent remarkable studies will be addressed by the three speakers, who have succeeded in revealing the mechanism of the photosystem II at the atomic and electronic levels after intensive international competition, using X-ray Free Electron Laser (XFEL) and the quantum chemistry.

講演者・プログラム：

1. 沈 建仁(岡山大学大学院自然科学研究科) Jian-Ren Shen (Okayama Univ.)
タイトル：立体構造解析から探る可視光を利用した水分解反応の仕組み
Mechanism of light-induced water-splitting reaction based on crystal structure analysis of photosystem II

概要 [Abstract]
緑色植物などの光合成は太陽の光エネルギーを利用して水と二酸化炭素から酸素と有機物を作ることで、地球上生物の生存を支えている。このうち、光エネルギーを利用して水を分解し、酸素、水素イオン、電子を放出する反応は光エネルギーの化学エネルギーへの変換、大気中への酸素の供給の点で重要なものである。この反応は光化学系 II 膜タンパク質複合体によって触媒されており、この巨大膜タンパク質の高分解能構造解析によって反応の仕組みが分かってきた。この反応をモデルとして利用することで、太陽からクリーンで「無尽蔵」なエネルギーを取りだすことができるかもしれない。

Oxygenic photosynthesis by green plants etc. is vital for survival of organisms on Earth by providing oxygen and organic substances from water and carbon dioxide utilizing light energy from the sun. Among photosynthetic reactions, the light-induced water-splitting reaction generates oxygen, protons and electrons, which therefore is important in terms of light-energy conversion and the production of oxygen. The water-splitting reaction is catalyzed by Photosystem II (PSII), a huge membrane-protein complex. Through high-resolution structural analysis of PSII, the mechanism of water-splitting is becoming uncovered. It is expected that clean and limitless energy may be obtained from the sun by utilization of the water-splitting reaction in artificial photosynthesis.

2. 山本 雅貴(理研 RSC) Masaki Yamamoto (RIKEN SPring-8 Center)
タイトル: SACLA による無損傷 X 線結晶構造解析
Determination of damage-free crystal structure of X-ray sensitive proteins using SACLA

概要 [Abstract]
放射線損傷の無い原子分解能での X 線結晶構造はタンパク質の機能解明に重要である。しかし、高輝度放射光でのクライオ結晶構造解析でも放射線損傷を完全に解決できていない。実際クライオ条件下でも、チトクロム酸化酵素(CcO)の活性中心や光化学系 II(PSII)の酸素発生中心の放射線損傷が報告されている。フェムト秒の X 線自由電子レーザー(XFEL)パルスは放射線損傷によりタンパク質が破壊される前に回折像を記録可能である。それにより、CcO や PSII の様な大きな結晶を利用したフェムト秒 X 線結晶構造(FSC)は巨大タンパク質について無損傷での高分解能 X 線結晶構造解析を可能にした。本講演では SACLA での FSC の現状と展望を発表する。

Damage-free X-ray crystal structure at atomic resolution is important for the elucidation of the protein functions. The cryo-crystallography at SR partly mitigates radiation damage, but it has not completely solved it. In fact even at a cryogenic temperature, radiation damages of the active site of Cytochrome c oxidase (CcO) and the oxygen evolving complex of Photosystem II (PSII) were reported. A femtosecond X-ray Free Electron Laser (XFEL) pulse provides a diffraction image before the protein is destroyed by radiation damage. Therefore the femtosecond crystallography (FSC) using large crystals, e.g. CcO and PSII, have made possible the high resolution analysis of radiation damage free crystal structure of huge protein. We will present the current status and the future prospects of FSC at SACLA.

3. 庄司 光男(筑波大学数理物質科学研究科) Mitsuo Shoji (Univ. of Tsukuba)
タイトル:光化学系 II の酸素発生中心の電子状態
Electronic structures of the oxygen evolving complex in photosystem II

概要 [Abstract]
光合成において水分子を酸化する反応は光化学系 II の酸素発生中心で行われている。反応は CaMn4O5 クラスターで触媒されるが、複雑な電子状態を持つ為に、その構造と電子状態(酸化状態)がこれまで大きな議論となってきた。本発表では CaMn4O5 の取りうる電子状態と構造変化について近年の実験事実と計算結果を比較しながら解説する。

The key chemical reaction of photosystem is carried out at the oxygen-evolving complex (OEC) in photosystem II. The OEC contains a catalytic center called CaMn4O5 cluster, and the geometrical structures and electronic structures (oxidation forms) have been the major debating topics. In our presentation, possible electronic structures and the structural changes of CaMn4O5 cluster are discussed with recent experimental findings and theoretical results.

4. 会場からの提案・質問

8. 理事会、会員総会、各種委員会の案内

委員会等	開催日程		会場
ホームページ編集委員会	9 月 12 日（土）	14:00 – 16:00	金沢大学サテライトプラザ
生物物理編集委員会		16:30 – 18:30	金沢大学サテライトプラザ
Biophysics and Physicobiology 編集委員会	9 月 13 日（日）	11:40 – 12:40	2 階 206 教室（諸会議室 1）
若手賞選考委員会		15:00 – 16:00	2 階 206 教室（諸会議室 1）
平成 27 年度第 2 回理事会（旧運営委員会）		18:30 – 19:30	図書館棟G1 階 G14 会議室（諸会議室 2）
企業との意見交換会	9 月 14 日（月）	9:30 – 11:00	2 階 206 教室（諸会議室 1）
男女共同参画・若手支援委員会		10:00 – 11:30	G2 階南福利食堂フレポ 奥（T 会場）
分野別専門委員会*1		11:30 – 12:20	1 階大講義室 A（H 会場）
会員総会（総会）*2		12:20– 13:40	1 階大講義室 A（H 会場）
若手の会会議		16:00 – 17:00	2 階 206 教室（諸会議室 1）

（ ）は法人化前の名称

*1; 分野別専門委員会では、英文誌「Biophysics and Physicobiology」セミナーが開催されます（3. 年会行事・プログラム）。

*2; 会員総会では、第 2 回会員総会ワークショップが開催されます（7. 第 2 回会員総会開催通知）。

謝 辞

本年会の開催・運営に当たり、以下の団体よりご協力・ご援助いただきました。
関係者一同より御礼申し上げます。

新学術領域研究「運動超分子マシナリーが織りなす調和と多様性」
新学術領域研究「温度を基軸とした生命現象の統合的理解」
新学術領域研究「少数性生物学－個と多数の狭間が織りなす生命現象の探求－」
新学術領域研究「新生鎖の生物学」
新学術領域研究「3D 活性サイト科学」
新学術領域研究「生命分子システムにおける動的秩序形成と高次機能発現」
新学術領域研究「動的クロマチン構造と機能」
新学術領域研究「動的構造生命科学が拓く新発想測定技術」
新学術領域研究「理論と実験の協奏による柔らかな分子系の機能の科学」
JST/CREST「ライフサイエンスの革新を目指した構造生命科学と先端的基盤技術」

第 53 回日本生物物理学会年会
実行委員長 安藤 敏夫

第53回日本生物物理学会年会 実行委員会

年会実行委員長　　安藤 敏夫（金沢大学理工研究域数物科学系）

実行委員

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渡邉 信嗣　（金沢大学理工研究域バイオAFM先端研究センター）

※50音順。敬称略。

■ Access

Access Information

■From Tokyo

JR Tokyo Sta. — Hokuriku Shinkansen Line (2.5 hours) — JR Kanazawa Sta. — Hokuriku Railroad Bus (35 min.) — Kanazawa University (Natural Science and Technology Main Hall)

Haneda Airport — 1hour — Komatsu Airport

- Komatsu Bus (12 min.) — JR Komatsu Sta. — Hokuriku Main Line (30 min.) — JR Kanazawa Sta.
- Hokuriku Railroad Bus Supper Express directly bound Kanazawa railway station (40 min.) — JR Kanazawa Sta.
- Hokuriku Railroad Bus Super Express bound for Kanazawa railway station through the downtown (40 min) — Korinbo — walk (5min) — Korinbo ①Bus Stop Shikokinenkan-mae — Hokuriku Railroad Bus (20 min.) — Kanazawa University

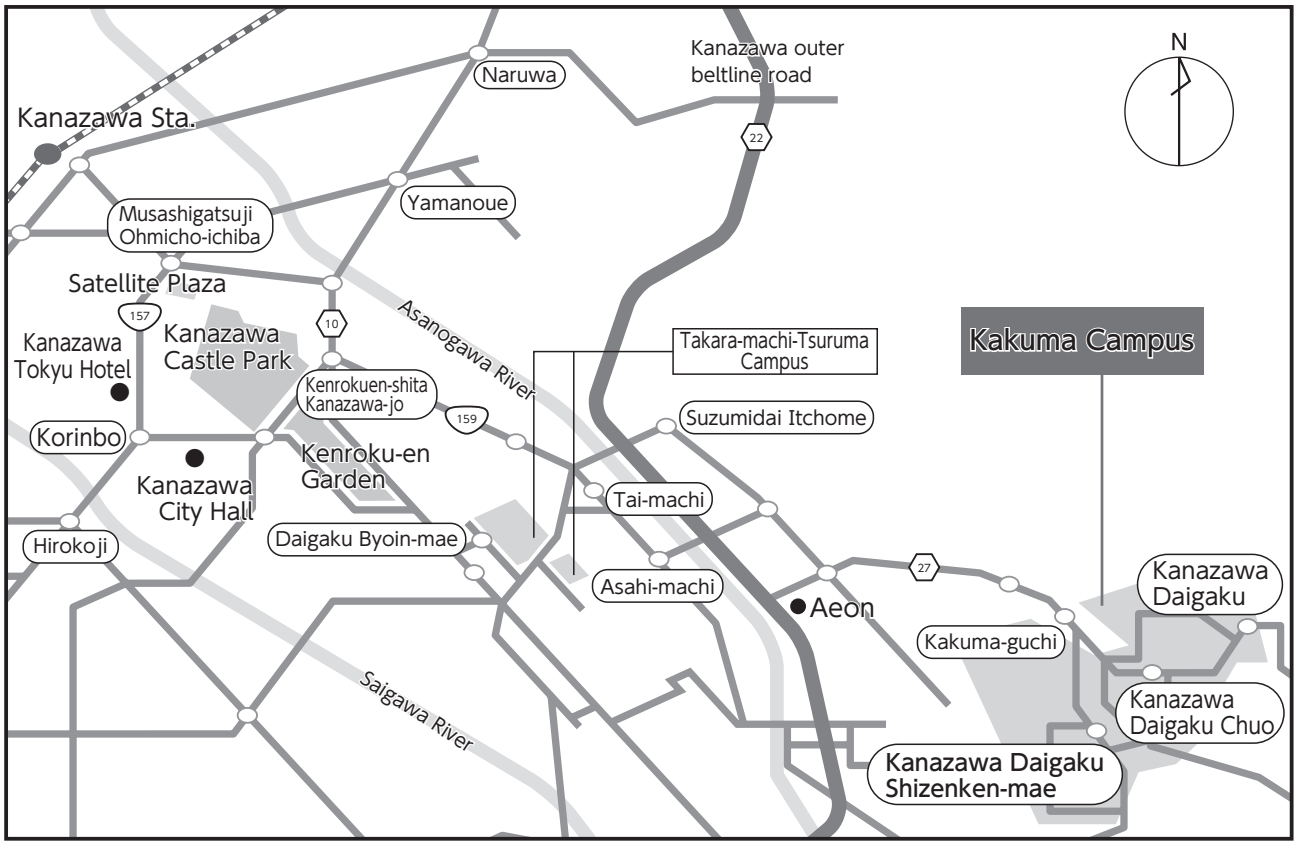
■From Osaka and Kyoto

JR Osaka Sta. / JR Kyoto Sta. — Limited Express. Thunderbird (2.5 hours) — JR Kanazawa Sta. — Hokuriku Railroad Bus (35 min.) — Kanazawa University

■From Nagoya

JR Nagoya Sta. — Limited Express Shirasagi (3 hours) — JR Kanazawa Sta. — Hokuriku Railroad Bus (35 min.) — Kanazawa University

Kanazawa city Map



Public bus service to Kanazawa University Kakuma campus (Hokuriku Railroad Co., Ltd. Route Bus)

Additional bus service will be provided during the meeting between Kanazawa Sta. and Kanazawa Daigaku Shizenken-mae (approx. 35 min).

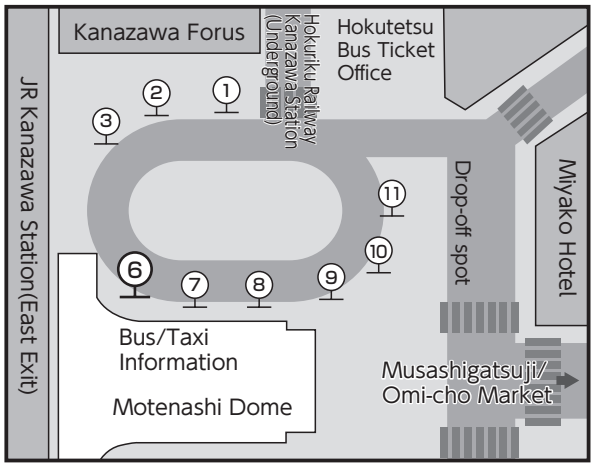
Date : 13 (Sun) －15 (Tue), September
Time : 7:50 - 8:30 (from Kanazawa Station)
After the end of each day's program (from Kanazawa Daigaku Shizennken Mae)
Fare : JPY360

■ From Kanazawa Sta. East Exit

Departs from No. 6 bus stop, Kanazawa Station east exit

Route:【91】(Weekdays only)
【93・94・97】Bound for Kanazawa Daigaku
Alighting bus stop: Kanazawa Daigaku Shizenken-mae (approx. 35 min)
Fare: JPY360

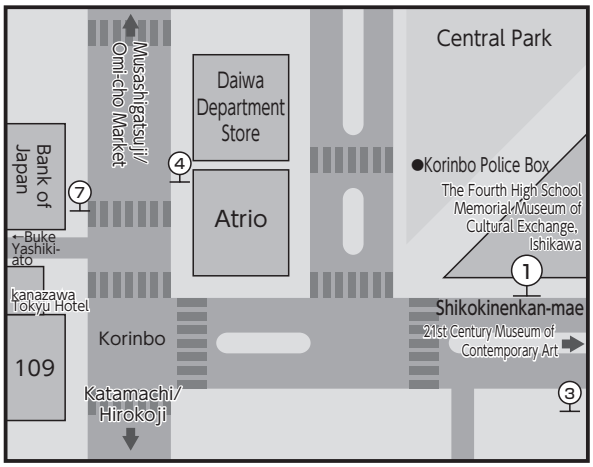
(By Taxi from Kanazawa Sta. to Kakuma campus: about JPY3,500)



■ From Korinbo

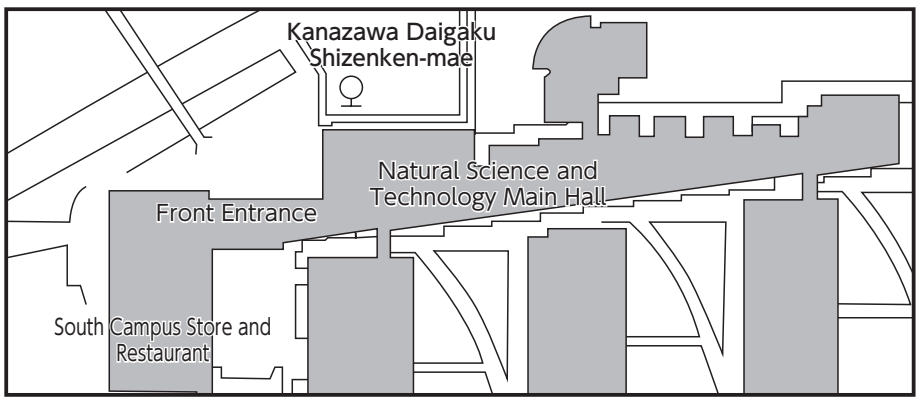
Departs from No.1 bus stop, Korinbo (Shikokinenkan-mae)

Route:【93・94・96・97】Bound for Kanazawa Daigaku
Alighting bus stop: Kanazawa Daigaku Shizenken-mae (approx. 20 min)
Fare: JPY360



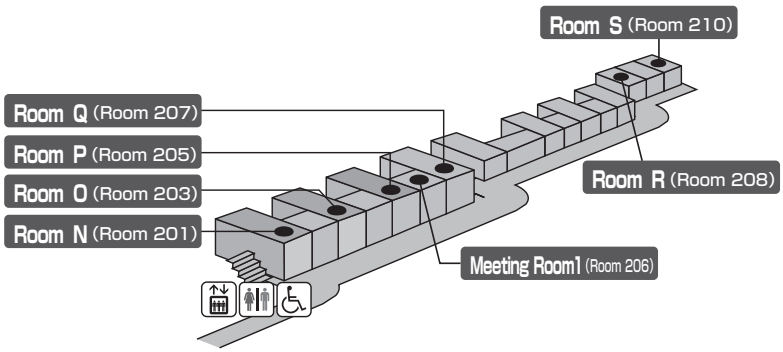
■ For Kanazawa Sta. East Exit

Additional bus service will be provided after the end of each day's program(approx 35 min).
Departs from Kanazawa Daigaku Shizenken-mae bus stop
Fare : JPY 360
Note : Don't board a bus bound for Kanazawa Daigaku.

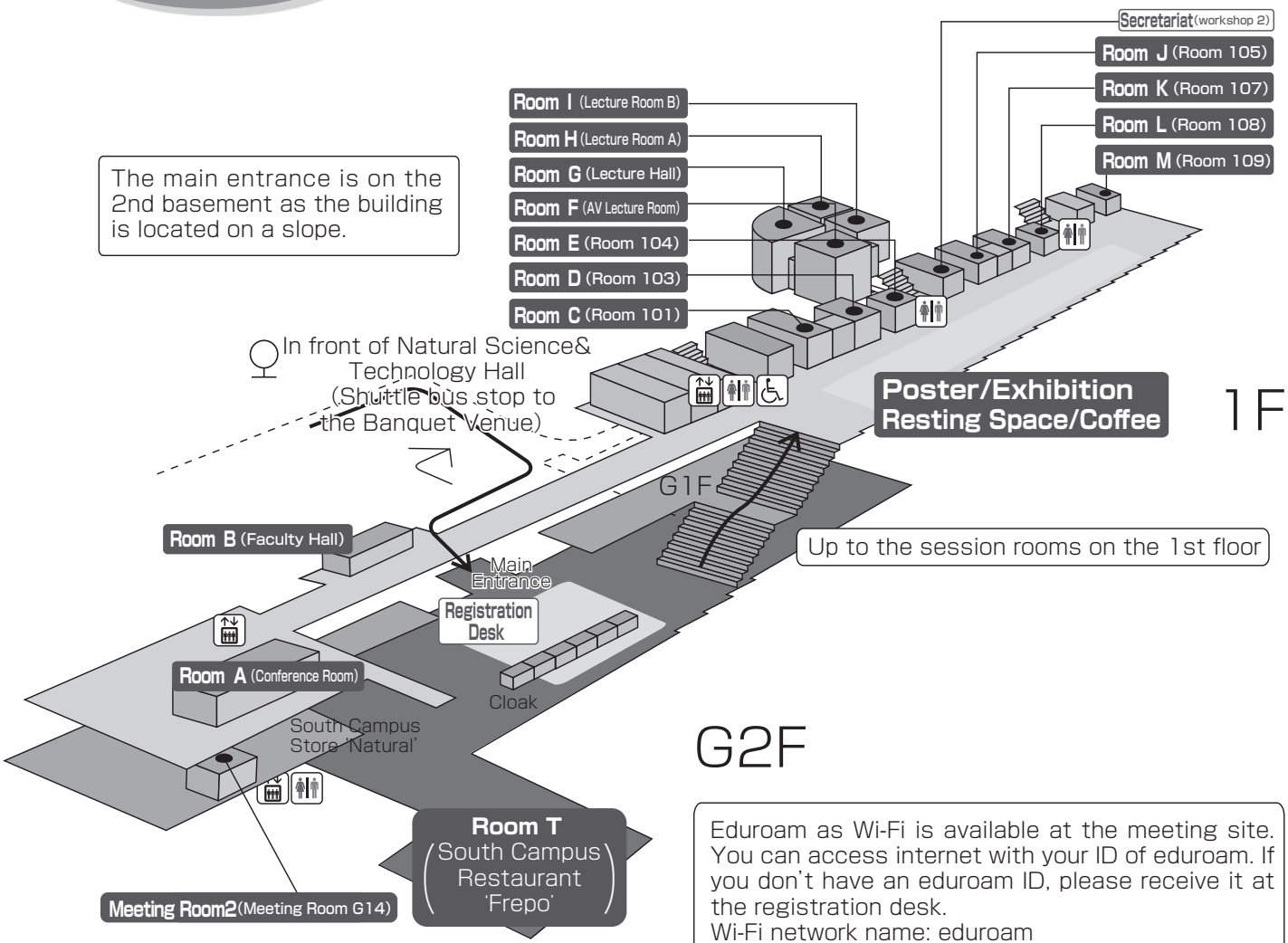


■ Floor Map (Natural Science and Technology Main Hall)

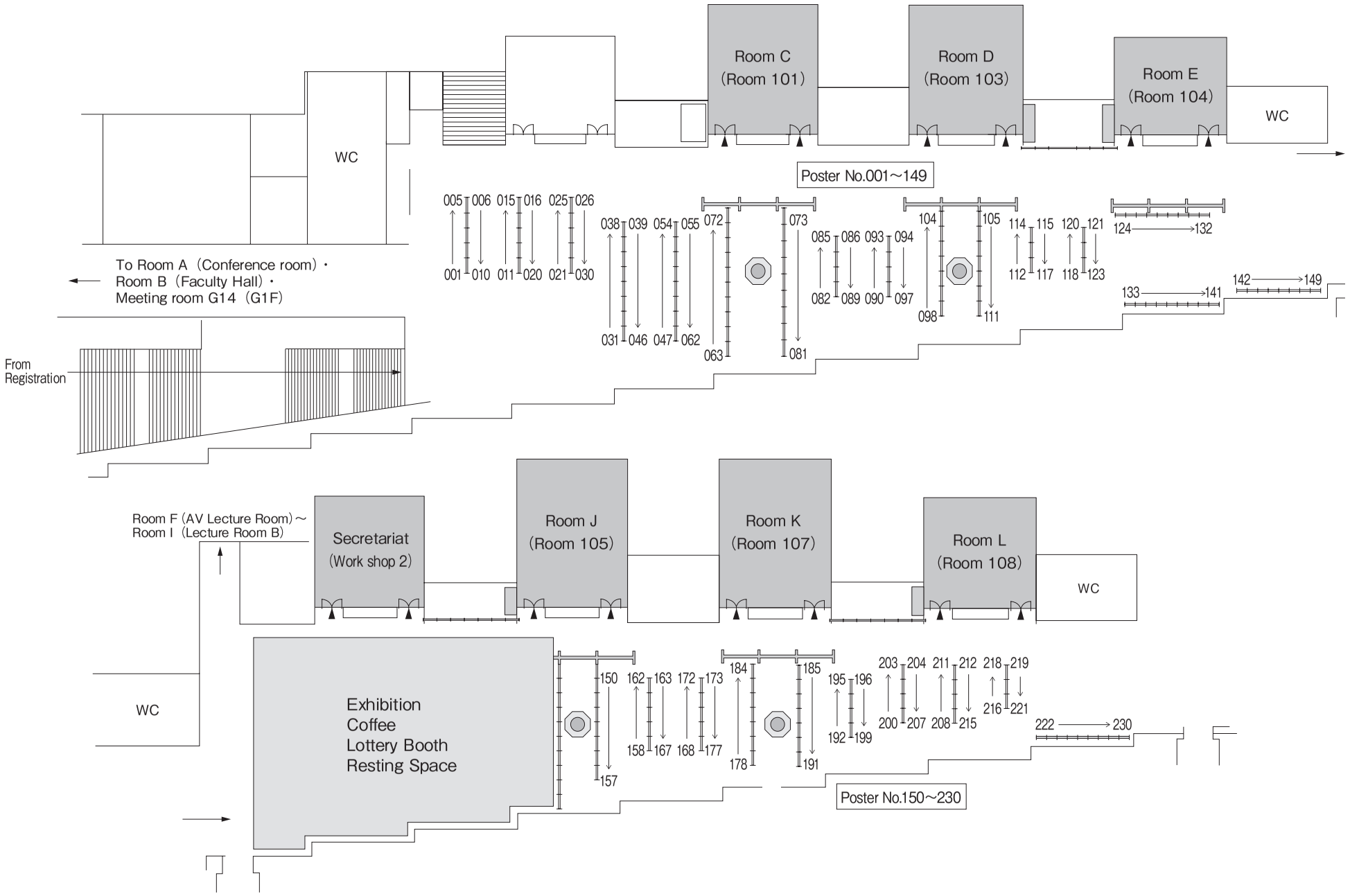
2F



1F/G1F/G2F



Eduroam as Wi-Fi is available at the meeting site. You can access internet with your ID of eduroam. If you don't have an eduroam ID, please receive it at the registration desk.
Wi-Fi network name: eduroam
※Wireless network can be unstable in some spots.
※Please refrain from using the internet during lectures.


Room C
(Room 101)
Room D
(Room 103)
Room E
(Room 104)
WC
WC
Poster No.001～149
To Room A (Conference room)・
Room B (Faculty Hall)・
Meeting room G14 (G1F)
From
Registration
Room F (AV Lecture Room)～
Room I (Lecture Room B)
Secretariat
(Work shop 2)
Room J
(Room 105)
Room K
(Room 107)
Room L
(Room 108)
WC
WC
Exhibition
Coffee
Lottery Booth
Resting Space
Poster No.150～230

September 13 (Sun.) Day 1

Japanese

Natural Science and Technology Main Hall, Kanazawa University Kakuma Campus

			9:00	10:00	11:00	12:00	13:00	14:00	15:00	16:00	17:00	18:00	19:00
1F	Conference Room	Room A	1SAA Diverse functions of flagella and cilia; –From molecules to mammalian development–					1SAP The dawn of thermal biology					
1F	Faculty Hall	Room B	1SBA Alkyne-tag based Raman probes and bio-imaging of small molecules			11:50	12:40	1SBP The challenge of experimentalists and theorists for the protein and nucleic acid system					
1F	Room 101	Room C	1SCA Novel 3D imaging of active sites in biomolecular systems to understand the biochemical functions			Career Support Joint Session (Luncheon Seminar) 11:50–12:40		1C Bioimaging I					
1F	Room 103	Room D	1SDA Large-scale networks in life science				The 4th Award Seminar of Outstanding Biophysics and Physicobiology Paper 12:50–13:20	1D Photobiology: Vision & Photoreception I					
1F	Room 104	Room E						1E Proteins: Structure, Structure-function relationship II		16:15			
1F	AV Lecture Room	Room F	"Early Career Award in Biophysics" Candidate Presentations			KAKENHI Guide (Luncheon Seminar)		1SFP Concerto and ensemble of membrane proteins and membrane lipids					
1F	Lecture Hall	Room G	1SGA Biosupercomputing opened by next-generation supercomputer post-K			1LG Hamamatsu Photonics K. K.		1SGP New developments in biophysical studies with X-ray free electron laser at the SACLA					
1F	Lecture Room A	Room H	1SHA Studies of dynamic chromatin structure and function to understand fundamentals of life			1LH Ryoka Systems Inc.		1SHP Conformational switching of tubulin and its physiological function					
1F	Lecture Room B	Room I	1SIA Future of biomimetics learning from natural history			1LI Protein Data Bank Japan (PDBj)		1SIP Principles and applications of super resolution microscopy in recent life science			16:20		
1F	Room 105	Room J	1SJA Advances in our understanding of the interaction between actin filaments and their binding proteins			11:50	12:40	1J Cell biology I					
1F	Room 107	Room K	1SKA Proton permeation mechanism across membrane					1K Molecular motor I					
1F	Room 108	Room L	9:00		11:30			1L Proteins: Function, Measurement, Analysis					
1F	Room 109	Room M						1M Proteins: Structure, Structure-function relationship I					
2F	Room 201	Room N						1Na Bioengineering 13:20–14:20		1Nb Membrane proteins I 14:25–16:15			
2F	Room 203	Room O						1O Nucleic acid binding proteins					
2F	Room 207	Room Q						1Qa Biological & Artificial membrane I 13:20–15:25		1Qb Nonequilibrium state & Biological rhythm 15:30–16:15			
2F	Room 210	Room S						Career Support Individual Session 13:20–14:32					
2F	Room 206	Meeting Room 1				Biophysics and Physicobiology Editorial Board Meeting 11:40–12:40	13:20		*ECAB Selection Meeting 15:00–16:00				
G1F	Meeting Room G14	Meeting Room 2			KAKENHI Guide Meeting 11:00–11:45								2015 2nd Board Meeting 18:30–19:30
1F	Promenade	Poster Session		Poster Setup, Display 8:45–16:30							Poster Presentation Odd num. 16:30–17:30	Poster Presentation Even num. 17:30–18:30	Removal
1F	Promenade	Exhibition				Exhibition 9:30–18:30							

*ECAB : Early Career Award in Biophysics

September 14 (Mon.) Day 2 Japanese

Natural Science and Technology Main Hall, Kanazawa University Kakuma Campus

			9:00	10:00	11:00	12:00	13:00	14:00	15:00	16:00	17:00	18:00	19:00
1F	Conference Room	Room A	2SAA Novel measurement techniques for visualizing 'live' protein molecules at work			2LA FEI Company Japan Ltd.		2SAP Principles and evolution for force generation in bio-nanomachines					
1F	Faculty Hall	Room B						2B Proteins: Structure, Structure-function relationship IV 13:55–16:00					
1F	Room 101	Room C	2Ca Molecular motor II 8:45–10:00					2Cb Bioimaging II		16:15			
1F	Room 103	Room D	2SDA Frontier of the study of lipid-active structures					2D Photobiology: Vision & Photoreception II					
1F	Room 104	Room E				General Assembly Workshop 12:20–13:40		2E Proteins: Property					
1F	AV Lecture Room	Room F	2SFA Description of 'living state' based on high-dimensional cellular dynamics			2LF Olympus Corporation	Career Support Joint Session 12:30–13:40	2SFP Advanced electron microscopy: A new world view of mesoscale biology					
1F	Lecture Hall	Room G	2SGA Functions of biomolecules controlled by flexibility			2LG Oxford Instruments		2SGP Dynamical structural change of proteins upon interaction with biomolecules					
1F	Lecture Room A	Room H	2SHA Biophysics underlying the synthesis and folding of nascent polypeptide chains			Experts Committee & Publication Ethics		2SHP Key role of pKa on functions of pump, enzyme, and motor					
1F	Lecture Room B	Room I	2SIA Formation of spatiotemporal dynamic ordering mediated by ATP hydrolysis			11:30	12:20	2SIP Mechanisms of diverged ligand recognition by similar protein structures for higher-order biological processes		16:25			
1F	Room 105	Room J	2SJA Synthetic biology exploring the border between life and material					2Ja Cell biology II 13:55–15:10	2Jb Heme proteins 15:15–16:15				
1F	Room 107	Room K	2SKA Diversity of GPCRs, and multimodal approach to understanding their functions					2K Proteins: Structure, Structure-function relationship III					
1F	Room 108	Room L	8:45		11:15			2L Bioinfomatics 13:55–15:10					
1F	Room 109	Room M						2Ma Muscle	2Mb Neuroscience 15:15–16:00				
2F	Room 201	Room N						2Na Membrane proteins II	2Nb Development & Differentiation				
2F	Room 203	Room O						2Oa Photobiology: Photosynthesis	2Ob Nucleic acid 15:15–16:15				
2F	Room 205	Room P						2P Mathematical biology					
2F	Room 207	Room Q						13:55 2Q Biological & Artificial membrane II					
2F	Room 210	Room S						Career Support Individual Session 14:00–16:00					
2F	Room 206	Meeting Room 1		Industry-Academia Discussion Meeting 9:30–11:00						Young Researchers Society Meeting 16:00–17:00			
1F	Cafeteria Frepo	Room T			Gender Equality Promotion & Young Researchers Committee Meeting 10:00–11:30	Gender Equality & Young Researchers Support Symposium 11:30–12:20							
1F	Promenade	Poster Session	Poster Setup, Display 8:30–16:30							Poster Presentation Odd num. 16:30–17:30	Poster Presentation Even num. 17:30–18:30	Removal	
1F	Promenade	Exhibition		Exhibition 9:30–18:30									

Banquet (KANAZAWA Tokyu Hotel) 19:30–21:30

September 15 (Tue.) Day 3

Japanese

Natural Science and Technology Main Hall, Kanazawa University Kakuma Campus

			9：00	10：00	11：00	12：00	13：00	14：00	15：00	16：00	17：00	18：00	19：00
1F	Conference Room	Room A	3SAA Frontiers of structural life science approaching to cellular phenomena										
	Faculty Hall	Room B	3SBA State-of-the-art hybrid methods for structural analysis of macromolecular complexes at functional state										
	Room 101	Room C	3SCA Electrophysiological approaches for structure and function of membrane proteins										
	Room 103	Room D	3SDA Dynamics of membrane transport proteins										
	Room 104	Room E	9：00			12：00							
	AV Lecture Room	Room F	3SFA New biophysical approaches to explore and manipulate cells				3LF OPTO-LINE, Inc.						
	Lecture Hall	Room G	3SGA Towards understanding origins of order through integrated approach of experiments and theory — From molecules to tissue —				3LG Rsearch Institute of Biomolecule Metrology Co., Ltd.						
	Lecture Room A	Room H	3SHA Biological events operated by small number of biomolecules				3LH NIKON INSTECH CO., LTD.						
	Lecture Room B	Room I	3SIA Protein structures and functions in multi-component systems: From hydration to intracellular environment				12：15 13：05						
	Room 105	Room J	3SJA Exploring rare events in biomolecular systems										
	Room 107	Room K	3SKA Artificial cells: Preparation, application, and activation										
	Promenade	Poster Session	Poster Setup, Display 8：45—13：30					Poster Presentation Odd num. 13：30—14：30	Poster Presentation Even num. 14：30—15：30	Removal			
		Exhibition	Exhibition 9：30—15：45										

Information for Participants and Presenters

1. Registration

◇ Registration desk

Location: Academic Hall (G2F), the entrance hall in the main building of Natural Science & Technology (Refer to the floor map page 28.)

Open Hours: Sep. 13(Sun) 8:30 – 17:00
14(Mon) 8:15 – 17:00
15(Tue) 8:30 – 15:00

◆Advance registration

For participants who have completed advance registration with full payment of the registration fee by the deadline, there is no need to stop by the registration desk. A name badge, a receipt and a program booklet have already been sent to these participants.

*Participants from overseas: Receive a name badge and a program booklet at the registration desk.
*Please wear your name badge throughout the meeting. Without it, you cannot enter the meeting site.
(Name badge holders will be provided at a place near the registration desk)

Note 1) Advance registration is completed only after the payment is done. In case your payment cannot be confirmed by the deadline, your registration is automatically cancelled. In this case, please register on-site at the registration desk.
Note 2) Name badges have not been sent to those who have not paid the BSJ annual membership fee. Please complete the payment at the BSJ desk of the meeting site.
Note 3) If you have already paid both registration and BSJ annual membership fees but not received a name badge, please visit the registration desk.
Note 4) For non-members who are invited to talk at a symposium, the membership is not required and your registration fee is waived. You are invited for free to the banquet.
Note 5) Attendees who belong to institutions outside of Japan can present at a general oral session or a poster session without membership.

◆On-site registration

Those who have not completed advance registration are required to register on-site at the registration desk. Only cash payment is acceptable.

◇ On-site registration fees

	BSJ Member				Non-Member		
	Regular	Senior	Student	Undergraduate student	Regular	Student	Undergraduate student
Registration fee	¥9,000	¥5,000	¥5,000	¥0	¥12,000	¥6,000	¥0
Banquet fee	¥7,000	¥5,000	¥5,000	¥3,000	¥7,000	¥5,000	¥3,000

・For undergraduate students, the registration fee is waived. You are required to present your student ID at the registration desk to receive a name badge and a program booklet. But the banquet fee is charged if you attend the banquet.

・For “Early Career Award in Biophysics” Candidate presenters, Biophysics and Physicobiology Outstanding Paper Award presenter and the representative of Biophysics and Physicobiology Editors’ Choice awardee, you are invited for free to the banquet. If you have already paid the banquet fee, you can get a full refund at the registration desk.

◇ Name badge

Please be sure to wear your name badge throughout the meeting. Entry without the badge is NOT acceptable. Remember to bring your name badge that was sent in advance. (Name badge holders will be provided at a place near the registration desk.)

◇ Receipt

A receipt is attached to the name badge. If you need another receipt form, it will be issued in exchange for the one attached to your name badge.

◇ Program booklet / Abstract online system【Release date: Aug. 18 (Tue)】

A program booklet (a part of front matters, and program) will be sent in advance to BSJ members and non-members with advance registration. The abstracts will be released only on the online system. No printed abstract booklet will be issued. On the online system, you can browse, search and download abstracts.
Program booklets can be purchased at the meeting site: JPY3,500/booklet.

Abstract online system: http://www.biophys.jp/dl/pro/53rd_proceedings.pdf Download ID: ambsj53 PW: kanazawa2015

The program (presentation title, presenter’s name and affiliation) and the online abstracts will be released on the BSJ53 web site. A half year later after the meeting, the abstracts will be posted on the CiNii web site which is linked from the BSJ web site.
BSJ web site: http://www.biophys.jp/ann/ann02.html
CiNii web site: http://ci.nii.ac.jp/organ/journal/INT1000001547_jp.html

◇ Program search system (Web version)【Release date: Aug. 18 (Tue)】

Program search system will be released on the BSJ53 web site.

◇ Free app of program search & abstracts browsing【Release date: Sep. 7 (Mon)】

The app is available for smart phones (iPhone /Android) and tablet computers (iPad /iPod Touch /Android). The app can be downloaded for free from App Store or Google Play.

App name:The 53rd Annual Meeting of the Biophysical Society of Japan Search word: bsj2015 PW of the abstracts browsing system:kanazawa2015

◇ BSJ membership (payment of the annual membership fee, and admission procedures)

For those who have not yet paid their annual membership fee, you can pay at the BSJ desk. For non-members, you are welcome to sign up at the BSJ desk to become a new member. For non-members who are invited to talk at a symposium or belong to institutions outside of Japan, the BSJ membership is not required.

2. Services & Facilities

◇ Cloakroom

Location: Academic Hall (G2F), the entrance hall in the main building of Natural Science & Technology
(Refer to the floor map page 28.)

Open Hours: Sep. 13 (Sun) 8:30 - 19:00
14 (Mon) 8:15 - 18:50
15 (Tue) 8:30 - 16:00

*Valuables or computers cannot be checked into the cloak since the society/meeting does not hold any responsibility for loss or damage of your items.

*Please pick your items up when you leave the meeting venue.

◇ Lunch

Free lunch:

Free lunch will be provided at luncheon seminars (day 1-3), KAKENHI Guide (day 1), and Gender Equality & Young Researchers Support Symposium (day 2). Lunch tickets will be distributed in the morning of day 1-3.

Please refer to luncheon seminar page.

Also a limited number of free lunch without lunch tickets will be provided at Experts Committee & Publication Ethics (day 2).

◆Restaurants & shop:

Restaurants & shop are available as follows.

- South campus restaurant "Frepo" (G2F)
  11:30 - 14:00 (last order 13:30; Sun, Mon, Tue)
  *Frepo will also open at 17:30 - 20:00 (only Mon & Tue).
- Restaurant "Sumire Tei"(Library building, 2F)
  11:00 - 14:30 (last order 14:00; Sun, Mon, Tue)
- Shop "Natural" (opposite side of Frepo) 9:00 - 17:00 (Sun, Mon, Tue)

◇ Paging service・bulletin board

No paging service is available to call an individual except for an emergency. Please use a bulletin board near the registration desk in order to contact with other participants.

◇ Parking

Parking lot is not available for participants.

◇ Accommodation

Please refer to "Accommodation" page of the BSJ2015 web site.

◇ Internet

Eduroam as Wi-Fi is available at the meeting site. You can access internet with your ID of eduroam. If you don't have an eduroam ID, please receive it at the registration desk.

Wi-Fi network name: eduroam

◇ Coffee

Coffee is available for free at the poster/exhibition place (1F promenade).

3. Programs & Events

*Several programs and events (committee meetings, general assembly meeting and its associated workshop, and lecture for public) are omitted here.

◇ "Early Career Award in Biophysics" Candidate Presentations

Date & Time: Sep.13 (Sun) 9:00 - 11:30
Place: Room F (AV Lecture Room)

◇ Lecture by Biophysics and Physicobiology Outstanding Paper Awardees

Date & Time: Sep. 13 (Sun) 12:50 - 13:20
Place: Room H (Lecture Room A)

◇ Banquet

Date & Time: Sep. 14 (Mon) 19:30 - 21:30
Place: KANAZAWA Tokyu Hotel 5F "The Ballroom" (One-minute walk from Korinbo bus stop)
Address: 2-1-1, Korinbo, Kanazawa city Tel: 076-231-2411
*For moving to the hotel, hired buses are available for free. Departure time: 18:45 - 19:00
*On-site registration is available at the registration desk or the banquet reception desk.

◇ Career Support Session

Date & Time: Sep. 13 (Sun) 10:00 - 11:40、13:20 - 16:20 (Details will be bulletined on a board)
14 (Mon) 9:00 - 11:20、14:00 - 16:00 (Details will be bulletined on a board)
Place: Room S (Room 210)
Who to attend: Students and researchers who are seeking for a job
Contents: Employer side (companies, research institutions and laboratories) will provide a brief introduction of their activities and jobs and then discuss with the attendees.

◇ Luncheon seminar

Lunch tickets will be distributed at the luncheon seminar desk, as shown below.

◆Distribution of lunch tickets

Luncheon seminar desk
Hours: Sep. 13 (Sun), 15 (Tue) 8:30 - 10:30 / 14 (Mon) 8:15 - 10:30
*The desk will be closed when all the tickets are distributed.
Location: Academic Hall (G2F), near the registration desk

*Only tickets for the seminars on the day are provided on a first-come first-served basis.
*Lunches are provided by courtesy of companies and groups co-sponsoring luncheon seminars.

◆Attention

Please note that the lunch tickets will become invalid when you do not come before the starting time of the seminars and that the resulting remaining lunches will be provided to those who are attending the seminars without lunch tickets.

◇ Exhibition

Instruments, reagents, software, books, etc. are displayed at the exhibition hall (1F promenade).

4. Prohibited Items

◇ Photography & recording

Photography and recording with camera, video, mobile phone and any device is NOT allowed at the meeting site.

◇ Smoking, drinking & eating

Smoking is NOT allowed at the meeting site except for the separate smoking spots. Drinking and eating is NOT allowed inside lecture rooms except for luncheon seminars and other seminars/meetings in which meals are served.

◇ Cell-phone use

Talking on a mobile phone in the lecture/presentation rooms is NOT allowed. Please set your mobile phone on the silent mode or off, and make sure it will not make noises during lectures/presentations.

5. Contact

◇ During the meeting

Secretariat (Tel: 07-5453-8365　*phone number reachable during the meeting)

◇ Before or after the meeting

The organizing committee of the BSJ53
53bp-nenkai@ml.kanazawa-u.ac.jp
Registration and abstract submission support desk
Nakanishi Printing Company, Tokyo Branch,
Hamada Bldg. 5F, 2-26-1, Hongo, Bunkyo-ku, Tokyo 113-0033
bsj2015sys-sprt@e-naf.jp
Support team, exhibition and advertisement secretariat
A & E planning Co., Ltd.
Iwanami Shoten Hitotsubashi Bekkan 4F, 2-4-4 Hitotsubashi, Chiyoda-ku, Tokyo 101-0003
Tel: 03-3230-2744 / Fax: 03-3230-2479
Support team: jbp2015@aeplan.co.jp
Exhibition and Advertisement secretariat: e_jbp2015@aeplan.co.jp

6. Information for Presenters

◇ Language

Prepare your slides in English and give your presentation in English.

◇ Projector

Please bring a laptop with you for your presentation. A projector is equipped in each lecture room.
A sound output is not accepted.

1) Please prepare your presentation file in Microsoft PowerPoint.
2) The output resolution should be XGA (1024 x 768). The higher resolutions would possibly lose some information.
3) Our staff will connect your laptop to a video switcher.

4) The video output connector of your laptop should be "miniD-sub15pin(female)". If your connector is a different type (for example, that of Macintosh computer), please bring a conversion adaptor.
5) Bring your PowerPoint file in a USB memory.
6) Bring your AC adaptor in case that your battery would die.
7) Deactivate the screen-saver and power saving mode of your laptop.

◇ For organizers of symposia & "Early Career Award in Biophysics" (ECAB) Candidate presentations

Please come to the assigned room by 15 minutes before the start of the session, and then tell our staff of your arrival. Keep the time schedule and make smooth progress in the program. As a time keeper, our staff will help you.

Time allocation:
Symposium: Time allocation will be controlled by chairpersons.
ECAB Candidate Presentation: Presentation 10min. + Discussion 3min. + Laptop change 2min.

◇ For speakers of symposia & ECAB Candidate presentations

Please come to the "PC Reception Desk" in the assigned room by 15 minutes before the start of the session. Our staff will connect your computer to a video switcher.
*Please note that there is no preview room.

ECAB Candidate Presentation: Presentation 10min. + Discussion 3min. + Laptop change 2min.

After ECAB Candidate presentations, a certificate of your presentation at this event will be granted to you in the room.

◇ For chairpersons of general oral sessions

Please come to the assigned room at least 10 min before the start of the assigned session, and then inform our staff of your arrival. Keep the time schedule and make smooth progress of the session. As a time keeper, our staff will help you.

Time allocation: Presentation 10min. + Discussion 4min. + Laptop change 1min.

◇ For speakers of general oral sessions

Please come to the PC Reception Desk in the assigned room before 20 min before your presentation. Our staff will connect your laptop to a video switcher. *Please note that there is no preview room.

Time allocation: Presentation 10min. + Discussion 4min. + Laptop change 1min.

◇ For poster presenters

		Day 1, Sep. 13	Day 2, Sep. 14	Day 3, Sep. 15
Setup, Display		8:45-16:30	8:30-16:30	8:45-13:30
Presentation Discussion	Odd Numbers	16:30-17:30	16:30-17:30	13:30-14:30
	Even Numbers	17:30-18:30	17:30-18:30	14:30-15:30
Removal		until 18:45	until 18:45	until 15:45

*Periods of poster display: Posters will be replaced every day for the next day's poster presentations.
*Panel size: 90cm wide x 210cm high. Push pins are available at the site.
*Removal: Any posters remaining on panels after the removal time will be discarded by the secretariat.

◇ Instructions for poster presentation

A poster must be written in English.
Put a small circle on the upper left of the presenter's name.

◇ Presentation types and how to read the presentation numbers

Presentation types are Symposium Speech, "Early Career Award in Biophysics" Candidate Presentations, Oral Presentation and Poster Presentation.

Speech at symposium: (Ex.) 1SAA-03
Presentation day (1) (1, Sep 13; 2, Sep 14; 3, Sep 15) + Symposium (S) + Session room (room A) + AM (A) / PM (P) + Order of the talk

"Early Career Award in Biophysics" Candidate Presentations: (Ex.) 1YF1045
Presentation day (1) (1, Sep 13; 2, Sep 14; 3, Sep 15) + Young Scientists (Y) + Session room (room F) + Starting time of the talk

Oral presentation: (Ex.) 1D1320
Presentation day (1) (1, Sep 13; 2, Sep 14; 3, Sep 15) + Session room (room D) + Starting time of the talk

Poster presentation: (Ex.) 1Pos001
Presentation day (1) (1, Sep 13; 2, Sep 14; 3, Sep 15) + Poster (Pos) + Panel number

第 53 回日本生物物理学会年会市民講演会
生物発光が拓く未来社会

日　時：9 月 12 日（土）開場：13 時 30 分、開演：14 時、終演：16 時

会　場：石川県教育会館 ホール

（〒 920-0961 金沢市香林坊 1-2-40　TEL: 076-222-1241）

JR 金沢駅よりバス 15 分（香林坊下車 / 徒歩 2 分），JR 金沢駅よりタクシー 12 分

参加費：無料（どなたでも自由に参加できます）

主　催：第 53 回日本生物物理学会年会実行委員会

世話人：古寺 哲幸（金沢大学理工研究域バイオＡＦＭ先端研究センター）

安藤 敏夫（金沢大学理工研究域数物科学系）

講演プログラム

「生物発光が拓く未来社会」

永井 健治 教授（大阪大学産業科学研究所）

地球上にはホタルをはじめヤコウタケやオワンクラゲ等々、無数の発光生物が存在することが知られています。多くの場合ルシフェラーゼと呼ばれる酵素タンパク質がルシフェリンと呼ばれる発光物質に酸素を結び付ける事で光ることが明らかになっています。今では、ルシフェラーゼを遺伝子工学技術で改変すると、仄かな光を煌めく光に変えることができます。また、光る色を変えることだってできるのです。このような技術を我々の生活に応用しない手はありません。本講演会では、私たちの研究室で進めている生物発光を応用した電力を必要としない次世代照明灯 LEP の開発について紹介し、30 年後の未来社会に皆さまを誘(いざな)いたいと思います。

講演は日本語で行われます。

一般社団法人日本生物物理学会　第 4 回 Biophysics and Physicobiology 論文賞受賞講演会
The 4th Award Seminar of Outstanding Biophysics and Physicobiology Paper

オーガナイザー：日本生物物理学会 Biophysics and Physicobiology 論文賞選考委員会

Organizers: Award committee for Outstanding Biophysics and Physicobiology Paper

日　時：9 月 13 日（日）12:50 ～ 13:20 ／ Sep. 13 Sun.

場　所：H 会場（大講義室 A）／ Room H (Lecture Room A)

形　式：講演会／ Lecture

第 4 回 Biophysics and Physicobiology 論文賞受賞者

BPPB Outstanding Paper Awardee

林 智彦[1]，松浦 東[2]，佐藤 博之[2]，櫻井 実[3]

Tomohiko Hayashi[1], Azuma Matsuura[2], Hiroyuki Sato[2], Minoru Sakurai[3]

[1] 京都大学エネルギー理工学研究所 [2] 富士通研究所 [3] 東京工業大学バイオ研究基盤支援総合センター

[1] Institute of Advanced Energy, Kyoto University [2] Fujitsu Laboratories, Ltd. [3] Center for Biological Resources and Informatics, Tokyo Institute of Technology

全原子量子化学計算による光活性タンパク質の吸収波長制御機構の解析

Analyses of the spectral-tuning mechanisms of several photoactive proteins based on the full-quantum chemical calculations

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The photoactive proteins, which absorb light and convert it into biological signal or chemical energy, consist of a chromophore and an apoprotein. The absorption spectra of each protein are finely tuned due to the chromophore-apoprotein interactions. Elucidation of this spectral-tuning mechanism has been a central issue in biophysics. A typical theoretical approach toward achieving such elucidation is that the protein of interest is regarded as a hybrid system consisting of quantum-mechanical (QM) and molecular-mechanical (MM) regions (QM/MM). In this approach, the chromophore alone or a small region involving the chromophore and its neighboring amino acid residues are treated using a QM method, and its surrounding is approximated using a classical MM method. However, QM/MM approaches have some ambiguity concerning how to partition the system into QM and MM regions. To overcome this ambiguity, we developed a "Full-QM" theoretical approach and successfully applied it to the absorption-maximum calculations of several photoactive proteins. In our BIOPHYSICS paper in 2012[1], we analyzed the spectral-tuning mechanism of bacteriorhodopsin (bR), and showed that an important factor contributing to the spectral tuning of bR is the electron transfer from the apoprotein to the chromophore upon light absorption.

[1] Hayashi, T., Matsuura, A., Sato, H. & Sakurai, M. *BIOPHYSICS*, 8, 115–125 (2012).

男女共同参画・若手支援委員会企画・グループディスカッション
『ポスドク問題』って言わないで！ー任期付き雇用問題の解決を目指してー
The Symposium for the Promotion of Gender Equality and Young Researchers
Don’t call it a “Post-Doc Problem”!
–Group discussion on the issue of a fixed-term employment–

オーガナイザー：日本生物物理学会　男女共同参画・若手支援委員会

Organizers: Promotion of Gender Equality and Young Researchers Committee

日　時：9 月 14 日（月）11:30 〜 12:20（ランチョンセミナーの時間帯）

会　場：T 会場（南福利食堂フレポ奥）

昼　食：お弁当とお茶を無料で提供いたします。ただし、数に限りがあります。

形　式：グループディスカッション（Japanese language will be used in the discussion.）

概要：本年 1 月、生物科学学会連合（生科連）のポスドク問題検討委員会は「生科連からの＜重要なお願い＞」と題した文書を文部科学省に提出しました。この文書では、1 万 6 千人（2012 年）を越えるとされる博士研究員の現状を分析し、博士研究員のキャリアパス創出や支援を目指したさまざまな提案がなされています。本グループディスカッションは、この問題に関心を持つ会員同士の議論を通して、学会として取り組むべき課題を探ると共に、立場の異なる会員間のネットワークを作る場として企画しました。

一般に「ポスドク問題」と呼ばれますが、ポスドクだけが問題なのではありません。むしろ、任期付き雇用が多く存在する一方で、常勤の研究職数が全く増えていないことこそが問題です（生科連文書でも、５年以下の任期で再任がない職をポスドクと定義しています)。これにより、研究者を目指す若者が減少する可能性も指摘されています。昨今では、ポスドクは「特任」や「テニュアトラック」などと呼称を変えつつありますが、将来設計が難しい職種であることには変わりません。さらには、信頼性の高いデータを出せるテクニシャンも、高度な技術が認められているにも関わらず、任期付きです。また、改正労働契約法への対応としての雇い止めの可能性も大変大きな問題と言えるでしょう。

本企画は、任期付き職全体にフォーカスし、問題点を共有すると共に、研究の活性化や新たなキャリアパス創成に向けた経験等を話し合う場にしたいと思います。現在まさにポスドクである方々だけではなく、この問題に関心をお持ちの多様なバックグラウンドの皆様の参加をお待ちしております。

「JSPS 特別研究員と科研費の制度：最近の動向」
Current Activities of JSPS Research Fellowship Program for Young Scientists and the Grants-in-Aid System

世話人：寺北 明久（大阪市立大学大学院理学研究科、日本学術振興会学術システム研究センター専門研究員）
Organizer：Akihisa Terakita (Graduate School of Science, Osaka City University; Program Officer, Research Center for Science Systems, JSPS)

日　時：9 月 13 日（日）11:50 ～ 12:40（ランチョンセミナーの時間帯）
会　場：F 会場（AV 講義室）
昼　食：お弁当とお茶が無料で提供されます。ただし、数に限りがあります。
形　式：講演会

○「科研費」の最近の動向

講師：日本学術振興会　研究助成第一課長　大鷲 正和

日本学術振興会から、科研費の配分機関として制度の概要、応募から審査、決定までの流れを中心に、補助金、助成金の執行と適切な管理、不正防止に関すること、また、成果の公開、科研費の普及啓発などについても、ご説明いただきます。

○特別研究員制度について

講師：日本学術振興会　人材育成事業部 研究者養成課　第二係長　林 崇宏

日本学術振興会人材育成事業部で行っている研究者養成事業（特別研究員、海外特別研究員、振興会賞、育志賞）の概略と学術システム研究センターの役割について、特別研究員制度を中心にご説明いただきます。また、審査方針から読み解く特別研究員として求められる資質、申請書作成時の心構えについてもご解説いただきます。

若手招待講演　Early Career Award in Biophysics

第 1 日目（9 月 13 日（日）／Day 1（Sep. 13 Sun.））

9:00～11:30　F 会場／Room F：AV 講義室/AV Lecture Room
1YF　日本生物物理学会若手奨励賞選考会
"Early Career Award in Biophysics" Candidate Presentations

オーガナイザー：若手奨励賞選考委員会
Organizer: Selection Committee for the Early Career Award in Biophysics

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In 2005, the Biophysical Society of Japan has established Early Career Award in Biophysics to recognize distinguished research work by young members of the BSJ. In this eleventh year, we received 39 highly qualified applications. After extremely competitive first round of screening based on written application forms, the following ten applicants were selected as the “young guest speakers.” For the second round of the nomination, each young speaker will be asked to make a 10-minute presentation followed by 3-minute Q&A discussion. At the end of these rounds, up to five award winners will be selected. The award winners will be announced at the banquet in the evening of Monday 14th September, and the winners will deliver a short talk. We welcome all the BSJ members to attend the oral presentations on Sunday 13th September at the Early Career Award in Biophysics Candidate Presentations and would like the members to foresee the future of biophysics in Japan through these speakers and their researches.

09:00　伊藤 創祐　1Q1600
1YF0900　フィードバックループのあるシグナル伝達におけるマックスウェルのデーモン
Maxwell's demon in biochemical signal transduction with feedback loop
○伊藤 創祐[1], 沙川 貴大[2]（[1]東京工業大学, 物性物理学専攻, [2]東京大学, 工学系研究科）
Sosuke Ito[1], Takahiro Sagawa[2] ([1]*Department of Physics, Tokyo Institute of Technology*, [2]*Department of Applied Physics, the University of Tokyo*)

09:15　加藤 英明　1M1335
1YF0915　光駆動性ナトリウムイオンポンプによるナトリウムイオン輸送の構造基盤
Structural basis for Na^+ transport mechanism by a light-driven Na^+ pump
○加藤 英明[1], 井上 圭一[2], 吉住 玲[2], 加藤 善隆[2], 大野 光[2], 今野 雅恵[2], 細島 頌子[3], 石塚 徹[3], Mohammad R. Hoque[3], 國友 博文[4], 伊藤 淳平[5], 吉澤 晋[6], 山下 恵太郎[7], 武本 瑞樹[4], 西澤 知宏[4], 谷口 怜哉[4], 木暮 一啓[6], Andres D. Maturana[5], 飯野 雄一[4], 八尾 寛[3], 石谷 隆一郎[4], 神取 秀樹[2], 濡木 理[4]（[1]スタンフォード大・医学, [2]名工大・院工, [3]東北大・院生命科, [4]東大・院理, [5]名大・院生命農, [6]東大・海洋研, [7]理研・播磨）
Hideaki Kato[1], Keiichi Inoue[2], Rei Yoshizumi[2], Yoshitaka Kato[2], Hikaru Ono[2], Masae Konno[2], Shoko Hososhima[3], Toru Ishizuka[3], Mohammad R. Hoque[3], Hirofumi Kunitomo[4], Jumpei Ito[5], Susumu Yoshizawa[6], Keitaro Yamashita[7], Mizuki Takemoto[4], Tomohiro Nishizawa[4], Reiya taniguchi[4], Kazuhiro Kogure[6], Andres D. Maturana[5], Yuichi Iino[4], Hiromu Yawo[3], Ryuichiro Ishitani[4], Hideki Kandori[2], Osamu Nureki[4] ([1]*Sch. of Med., Stanford Univ.*, [2]*Grad. Sch. of Engineering, Nagoya Inst. of Tech.*, [3]*Grad. Sch. of Life Sci., Tohoku Univ.*, [4]*Grad. Sch. of Sci., Univ. of Tokyo*, [5]*Grad. Sch. of Bioagri. Sci., Nagoya Univ.*, [6]*Atmos. and Ocean Res. Inst., Univ. of Tokyo*, [7]*Harima Inst., Riken SPring-8*)

09:30　黒井 邦巧　1L1320
1YF0930　光センサータンパク質 TePixD の反応過程における過渡的揺らぎ
Transient conformational fluctuation of TePixD during a reaction
○黒井 邦巧[1], 岡島 公司[2,3], 池内 昌彦[2], 徳富 哲[3], 寺嶋 正秀[4]（[1]分子研, [2]東大院・理, [3]大阪府大院・理, [4]京大院・理）
Kunisato Kuroi[1], Koji Okajima[2,3], Masahiko Ikeuchi[2], Satoru Tokutomi[3], Masahide Terazima[4] ([1]*Inst. for Mol. Sci.*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*Grad. Sch. Sci., Univ. Osaka Pref.*, [4]*Grad. Sch. Sci., Univ. Kyoto*)

09:45　菅 倫寛　1M1405
1YF0945　X 線自由電子レーザーによって明らかにされた光化学系 II 複合体の 1.95Å 分解能での無損傷構造
Radiation damage free structure of oxygen evolving photosytem II at 1.95Å resolution revealed by X-ray Free Electron Laser
○菅 倫寛[1], 秋田 総理[1], 平田 邦生[2], 上野 剛[2], 村上 博則[2], 中島 芳樹[1], 清水 哲哉[1], 山下 恵太郎[2], 山本 雅貴[2], 吾郷 日出夫[2], 沈 建仁[1]（[1]岡山大, [2]播磨理研）
Michihiro Suga[1], Fusamichi Akita[1], Kunio Hirata[2], Go Ueno[2], Hironori Murakami[2], Yoshiki Nakajima[1], Tetsuya Shimizu[1], Keitaro Yamashita[2], Masaki Yamamoto[2], Hideo Ago[2], Jian-Ren Shen[1] ([1]*Okayama Univ.*, [2]*Riken Harima*)

10:00　高橋 康史　1C1455

1YF1000　ナノスケールの形状・化学物質濃度プロファイルを可視化するナノ電気化学顕微鏡の創成

Development of Nano Electrochemical Microscopy for Visualizing Nanoscale Cell Surface Topography and Chemical Profile

○高橋 康史[1,2,3], 井田 大貴[2], 珠玖 仁[2], 末永 智一[1,2]（[1]東北大WPI－AIMR, [2]東北大院環境, [3]JST さきがけ）

Yasufumi Takahashi[1,2,3], Hiroki Ida[2], Hitoshi Shiku[2], Tomokazu Matsue[1,2] ([1]*WPI-AIMR of Tohoku University*, [2]*Environmental studies, Graduate school of Tohoku University*, [3]*JST PREST*)

10:15　豊田 正嗣　2Q1355

1YF1015　植物の傷害応答性・長距離・高速カルシウムシグナル伝達

Mechanical wounding/insect attack-induced, long-distance, rapid calcium signal transduction in plants

○豊田 正嗣[1,2], Simon Gilroy[1]（[1]University of Wisconsin-Madison, [2]JST・さきがけ）

Masatsugu Toyota[1,2], Simon Gilroy[1] ([1]*University of Wisconsin-Madison*, [2]*JST, PRESTO*)

10:30　中村 修一　2Pos114

1YF1030　高分解能ステップ計測で分かってきた細菌べん毛モーターのサーマルラチェット機構

Thermal ratchet mechanism of the bacterial flagellar motor emerged by high-resolution nanophotometry

○中村 修一[1,2], 森本 雄祐[2,3], 上池 伸徳[2], 曽和 義幸[4], 南野 徹[2], 難波 啓一[2]（[1]東北大・院工, [2]阪大・院生命機能, [3]理研・QBiC, [4]法政大・生命科学）

Shuichi Nakamura[1,2], Yusuke V. Morimoto[2,3], Nobunori Kami-ike[2], Yoshiyuki Sowa[4], Tohru Minamino[2], Keiichi Namba[2] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*QBiC, RIKEN*, [4]*Dept. Frontier Biosci., Hosei Univ.*)

10:45　服部 峰充　1E1545

1YF1045　抗体の新たな抗原認識機構がもたらす特異性創出原理

New paradigm for antibody-antigen recognition enabling extraordinarily high specificity

○服部 峰充[1], Darson Lai[1], Irina Dementieva[1], Sherwin Montano[1], Kohei Kurosawa[1], Akiko Koide[1], Alexander J. Ruthenburg[1,2], 小出 昌平[1]（[1]Dept. of Biochem. and Mol. Biol., The Univ. of Chicago, [2]Dept. of Mol. Genetics and Cell Biol., The Univ. of Chicago）

Takamitsu Hattori[1], Darson Lai[1], Irina Dementieva[1], Sherwin Montano[1], Kohei Kurosawa[1], Akiko Koide[1], Alexander J. Ruthenburg[1,2], Shohei Koide[1] ([1]*Dept. of Biochem. and Mol. Biol., The Univ. of Chicago*, [2]*Dept. of Mol. Genetics and Cell Biol., The Univ. of Chicago*)

11:00　宮崎 牧人　3Pos127

1YF1100　細胞サイズ球状閉鎖空間内でのアクトミオシンリングの自発形成と収縮

Cell-sized spherical confinement induces the spontaneous formation of contractile actomyosin rings *in vitro*

○宮崎 牧人[1], 千葉 雅隆[1], 江口 宙輝[1], 大木 高志[1], 石渡 信一[1,2]（[1]早稲田大・物理, [2]早稲田バイオサイエンスシンガポール研究所）

Makito Miyazaki[1], Masataka Chiba[1], Hiroki Eguchi[1], Takashi Ohki[1], Shin'ichi Ishiwata[1,2] ([1]*Dept. of Physics, Waseda Univ.*, [2]*WABIOS, Waseda Univ.*)

11:15　森本 大智　1Pos040

1YF1115　ユビキチン化の新規物理化学的性質

Novel physicochemical properties of ubiquitylation

○森本 大智[1], Erik Walinda[1], 菅瀬 謙治[1], 深田 はるみ[2], 曽 友深[3], 蔭山 俊[3,4], 星野 大[5], 藤井 高志[6], 土屋 光[7], 佐伯 泰[7], 有田 恭平[8], 有吉 眞理子[1], 杤尾 豪人[9], 岩井 一宏[10], 難波 啓一[6,11], 小松 雅明[3,4], 田中 啓二[7], 白川 昌宏[1]（[1]京大・工, [2]大府大・生命環境, [3]東京都医学研・蛋白質リサイクル, [4]新潟大・医, [5]京都大・薬, [6]理研・QBiC, [7]東京都医学研・蛋白質代謝, [8]横市大・生命医, [9]京都大・理, [10]京都大・医, [11]大阪大・生命機能）

Daichi Morimoto[1], Erik Walinda[1], Kenji Sugase[1], Harumi Fukada[2], Yu-shin Sou[3], Shun Kageyama[3,4], Masaru Hoshino[5], Takashi Fujii[6], Hikaru Tsuchiya[7], Yasushi Saeki[7], Kyohei Arita[8], Mariko Ariyoshi[1], Hidehito Tochio[9], Kazuhiro Iwai[10], Keiichi Namba[6,11], Masaaki Komatsu[3,4], Keiji Tanaka[7], Masahiro Shirakawa[1] ([1]*Eng., Kyoto Univ.*, [2]*Life Envi. Sci., Osaka Pref. Univ.*, [3]*Protein Metabolism Proj., Tokyo Metro. Ins. Med. Sci.*, [4]*Med., Niigata Univ.*, [5]*Pharm., Kyoto Univ.*, [6]*QBiC, RIKEN*, [7]*Lab. Protein Metabolism, Tokyo Metro. Ins. Med. Sci.*, [8]*Med. Life Sci., Yokohama City Univ.*, [9]*Science, Kyoto Univ.*, [10]*Med., Kyoto Univ.*, [11]*Frontier Biosci., Osaka Univ.*)

第 1 日目（9 月 13 日（日））／Day 1（Sep. 13 Sun.）

9:00～11:30　A 会場（大会議室）／Room A (Conference Room)

1SAA　べん毛・繊毛が織りなす多様な生命現象に挑む　～分子から個体まで～
Diverse functions of flagella and cilia; –From molecules to mammalian development–

オーガナイザー：西坂 崇之（学習院大学），池上 浩司（浜松医科大学）
Organizers: Takayuki Nishizaka (Gakushuin University), Koji Ikegami (Hamamatsu University School of Medicine)

Flagella and cilia, which behave as 'extracellular' organelles, are involved in a huge variety of biological processes, while having evolutionally-conserved common structures. In this symposium, ingenious researchers present latest and fascinating studies that cover a wide range of topics of living organisms, from prokaryotes to mammals, to reveal the biological roles of flagella and cilia. We would offer the audience an opportunity to learn creative and individual approaches from molecular levels to behavior levels.

1SAA-01　細菌べん毛モーターの力学特性
Dynamics of the nano-rotary motor of bacterial flagella
○曽和 義幸（法政大学生命科学部生命機能学科）
Yoshiyuki Sowa (*Dept. of Frontier Bioscience, Hosei Univ.*)

1SAA-02　先進光学顕微鏡により明らかになる繊毛とアーキアべん毛の力学機能
Mechanical function of cilia and archaeal flagella resolved by advanced optical microscopes
○西坂 崇之（学習院大学）
Takayuki Nishizaka (*Gakushuin Univ.*)

1SAA-03　Regulation of mammalian flagellar and ciliary motility by post-translational modifications of axonemal microtubule
Koji Ikegami, Mitsutoshi Setou (*Hamamatsu Univ. Sch. Med.*)

1SAA-04　マウス胚ノード繊毛においては中心構造体の欠損が回転運動パターンを可能とさせるがその代償として構造配置の不安定性をまねく
The absence of radial spokes allows rotational movement but confers ultrastructural instability in mouse node cilia
○篠原 恭介[1], Chen Duanduan[2], 西田 倫希[1], 美崎 佳寿代[3], 米村 重信[3], 浜田 博司[1]（[1]大阪大学, [2]北京理工大学, [3]理化学研究所）
Kyosuke Shinohara[1], Duanduan Chen[2], Tomoki Nishida[1], Kazuyo Misaki[3], Shigenobu Yonemura[3], Hiroshi Hamada[1] ([1]*Osaka University*, [2]*Bejing Institute of Technology*, [3]*RIKEN*)

1SAA-05　鞭毛繊毛ダイニンの多様性と運動の可変性
Diversity of axonemal dyneins and variability of their motor properties in different experimental systems
○神谷 律（学習院大・理・生命科学）
Ritsu Kamiya (*Department of Life Science, Gakushuin Univ.*)

1SAA-06　Shifting Gears with the Geometric Clutch
Charles B. Lindemann (*Oakland University, Department of Biological Sciences*)

9:00〜11:30　B 会場（ファカルティホール）／Room B (Faculty Hall)

1SBA　アルキンタグ等ラマンプローブをはじめとする小分子バイオイメージング最前線
Alkyne-tag based Raman probes and bio-imaging of small molecules

オーガナイザー：盛田 伸一（東北大学），石垣 美歌（関西学院大学）
Organizers: Shin-ichi Morita (Tohoku University), Mika Ishigaki (Kwansei Gakuin University)

Fluorescent tags are based on approximately 20 carbons, by which smaller molecules are impossible to be labelled. Recently, alkynes were attached to smaller molecules, which were successfully visualized within a living cell using a Raman microscope. The finding probably opened the new door in biophysics. Several years passed since then. What is the current state? In the meeting, the outstanding researchers are going to give talks about the latest results.

1SBA-01　Raman analysis of live cells with and without chemical markers
Shin-ichi Morita (*Tohoku Univ.*)

1SBA-02　１細胞解析のための発光センサーと光制御法の開発
Luminescent sensors and optical switches for single cell analysis
○小澤 岳昌（東京大学）
Takeaki Ozawa (*The University of Tokyo*)

1SBA-03　ラマン分光法を用いたマウス受精卵の分析
Analysis of mouse embryo by Raman spectroscopy
○石垣 美歌[1], 橋本 剛佑[2], 森本 香奈[1], 小川 直也[2], 尾崎 幸洋[1], 佐藤 英俊[2]（[1]関学理工化, [2]関学理工生命）
Mika Ishigaki[1], Kosuke Hashimoto[2], Kana Morimoto[1], Naoya Ogawa[2], Yukihiro Ozaki[1], Hidetoshi Sato[2] ([1]*Sci. and Tec. Chem., Kwan. Gak. Univ.*, [2]*Sci. and Tec. Biosci., Kwan. Gak. Univ.*)

1SBA-04　アルキン標識を用いた小分子のバイオラマンイメージング
Alkyne-tag Raman imaging of small molecules in biological systems
○安藤 潤[1,2,3]（[1]大阪大学・応用物理学専攻, [2]理化学研究所, [3]日本医療研究開発機構・AMED-CREST）
Jun Ando[1,2,3] ([1]*Dept. of Applied Physics, Osaka Univ.*, [2]*RIKEN*, [3]*AMED-CREST, AMED*)

1SBA-05　ラマンイメージングのための化学的に活性化可能なアルキンタグの開発
Chemically-activatable alkyne tag for Raman imaging
○山口 哲志（東大先端研）
Satoshi Yamaguchi (*RCAST, The Univ. of Tokyo*)

9:00〜11:30　C 会場（101 教室）／Room C (Room 101)

1SCA　新学術領域研究「３Ｄ活性サイト科学」共催
生体分子活性サイトの新しい３Ｄ構造決定法と機能解析
Novel 3D imaging of active sites in biomolecular systems to understand the biochemical functions

オーガナイザー：鷹野 優（広島市立大学），佐々木 裕次（東京大学）
Organizers: Yu Takano (Hiroshima City University), Yuji Sasaki (The University of Tokyo)

To understand biochemical functions such as photosynthesis, it is essential to determine accurate structures and dynamics of the active site in biomolecular systems. In materials science, atomic resolution holography and electron diffraction imaging are powerful methods to visualize 3D atomic images around the active site and is applying to visualization of the biological active site. In this symposium, we discuss how the collaboration of these novel 3D imaging methods and computer simulations contribute to the elucidation of the biochemical functions.

1SCA-01　原子分解能ホログラフィーによる生体分子活性サイトの３Ｄ原子構造直接解プロジェクト
Direct 3D atomic structure analysis project for active-site of bio-molecules by atomic-resolution holography
○大門 寛（奈良先端大）
Hiroshi Daimon (*Nara Inst. Sci. Tech. (NAIST)*)

1SCA-02 量子ビームを用いた 1 分子内部動態と活性サイト機能解析
Single Molecule Observations by Quantum Beams and Analysis of Functional Active-sites
○佐々木 裕次（東京大学大学院 新領域創成科学研究科 物質系専攻）
Yuji Sasaki (*Department of Advanced Materials Science, Graduate School of Frontier Sciences, The University of Tokyo*)

1SCA-03 アセチルコリン受容体のリガンド依存的チャネル開閉機構の解明に向けて ～X 線一分子計測と原子分解能ホログラフィーの可能性～
Toward understanding ligand-gated ion channels -Potential of diffracted X-ray tracking and atomic resolution holography-
○西野 有里[1], 関口 博史[2], 佐々木 裕次[3], 宮澤 淳夫[1]（[1]兵県大・院生命理学, [2]高輝度光科学研究センター, [3]東京大・院新領域創成科学）
Yuri Nishino[1], Hiroshi Sekiguchi[2], Yuji C. Sasaki[3], Atsuo Miyazawa[1] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*JASRI/SPring-8*, [3]*Grad. Sch. Frontier Sci., Univ. Tokyo*)

1SCA-04 ニトリル水和酵素の触媒機構に関する理論的研究
A QM/MM study of catalytic mechanism of nitrile hydratase
○栢沼 愛[1], 庄司 光男[1,2], 重田 育照[1,2]（[1]筑波大・計算セ, [2]筑波大・数理物質）
Megumi Kayanuma[1], Mitsuo Shoji[1,2], Yasuteru Shigeta[1,2] ([1]*Center of Comp. Sci., Univ. Tsukuba*, [2]*Grad. Sch. of Pure and App. Sci., Univ. Tsukuba*)

1SCA-05 生体試料に向けた蛍光 X 線ホログラフィーの挑戦
Challenge of X-ray fluorescence holography toward biomaterials
○林 好一（東北大学 金属材料研究所）
Koichi Hayashi (*Institute for Materials Research, Tohoku University*)

1SCA-06 ヘモグロビンのリガンド光解離過程における中間構造ダイナミクス観測
Structural dynamics measurements of the intermediate states in the ligand-photolysis of hemoglobin
○佐藤 文菜（自治医大・医）
Ayana Sato (*Sch. Med., Univ. Jichi*)

1SCA-07 ヘムの構造歪みの電子構造への影響に関する理論的研究
Theoretical study of the relationship between heme distortion and redox potential
○鷹野 優[1,2], 今田 康博[2]（[1]広島市大情報, [2]阪大蛋白研）
Yu Takano[1,2], Yasuhiro Imada[2] ([1]*Grad. Sch. Info. Sci., Hiroshima City Univ.*, [2]*IPR, Osaka Univ.*)

Closing remarks

9:00～11:30　D 会場（103 教室）／Room D (Room 103)

1SDA　生命科学における大規模ネットワーク解析
Large-scale networks in life science

オーガナイザー：黒田 真也（東京大学）, 岡田 眞理子（理化学研究所）
Organizers: Shinya Kuroda (The University of Tokyo), Mariko Okada (RIKEN)

Living systems are composed of a large-scale molecular networks across genome, transcriptome, proteome and metabolome. To elucidate the mechanisms in living systems, technologies for multi-omic measurements have recently been advanced, however, analysis of such a large-scale multi-omc data has yet to be developed.
In this symposium, recent advances in analysis of a large-scale networks will be introduced and discussed.

1SDA-01 マルチオミクスデータからのインスリン作用の大規模トランスオミクスネットワークの再構築
Reconstruction of a large-scale transomic network of acute insulin action from multi-omics data
○黒田 真也, 柚木 克之（東京大学理学系研究科生物科学専攻）
Shinya Kuroda, Katsuyuki Yugi (*Biological Sciences, University of Tokyo*)

1SDA-02 Trans-Omics analysis of the central carbon metabolism in Saccharomyces cerevisiae
Fumio Matsuda, Hiroshi Shimizu (*Grad. Sch. Inform. Sci. Tech., Osaka Univ.*)

1SDA-03 タンパク質相互作用を利用した統計的全ゲノム相関解析
Statistical assessment for genome-wide association study with protein-protein interactions
○瀬々 潤（産総研・創薬基盤）
Jun Sese (*BRD, AIST*)

1SDA-04 NF-κB pathway model の安定性・分岐解析
Stability and Bifurcation Analysis of an NF-κB pathway model
○田中 剛平（東京大学）
Gouhei Tanaka (*The University of Tokyo*)

1SDA-05 Dynamic behaviors of biochemical network
Mariko Okada (*RIKEN Center for Integrated Medical Sciences*)

9:00〜11:30　G 会場（レクチャーホール）／Room G (Lecture Hall)
1SGA　次世代スパコン「ポスト京」が拓くバイオスーパーコンピューティング
Biosupercomputing opened by next-generation supercomputer post-K

オーガナイザー：池口 満徳 （横浜市立大学）
Organizer: Mitsunori Ikeguchi (Yokohama City University)

Supercomputers continue to grow in scale and capability after the K computer in the world. In Japan, the development of the post K computer has been launched, and the huge computation power of the post K computer will be expected to push the boundary further in computational life sciences such as biomolecular simulations, genomic analysis, drug discovery, and health care. In this symposium, the future perspectives of biophysical supercomputing in the post-K-computer era will be discussed.

1SGA-01 スーパーコンピュータで明らかにするがんのヘテロ性
Unraveling cancer heterogeneity with supercomputer
○宮野 悟（東京大学）
Satoru Miyano (*The University of Tokyo*)

1SGA-02 循環器系の階層統合シミュレーション
Hierarchical Integrated Simulations of Circulatory System
○高木 周（東京大学）
Shu Takagi (*The University of Tokyo*)

1SGA-03 個別化医療支援に向けたデータ同化生体力学シミュレーション
Biomechanical simulation integrated with clinical measurements toward personalized medicine
○和田 成生（大阪大学 大学院基礎工学研究科）
Shigeo Wada (*Graduate School of Engineering Science, Osaka University*)

1SGA-04 ポスト「京」重点課題 1：生体分子システムの機能制御による革新的創薬基盤の構築
Innovative drug discovery infrastructure through functional control of biomolecular systems by using post 'K' supercomputer
○奥野 恭史[1,2]（[1]理化学研究所　生命システム研究センター, [2]京都大学　医学研究科）
Yasushi Okuno[1,2] ([1]*RIKEN Quantitative Biology Center*, [2]*Graduate School of Medicine, Kyoto University*)

1SGA-05 Large-Scale Molecular Simulation of Viruses: Multi-scale Molecular Modeling Approach
Wataru Shinoda, Kazushi Fujimoto, Yoshimichi Andoh, Susumu Okazaki (*Grad. Sch. Eng., Nagoya Univ.*)

1SGA-06 多剤排出トランスポーター AcrB の動的構造変化
Conformational Dynamics of Multidrug Efflux Transporter AcrB
○池口 満徳（横浜市大・院生命医）
Mitsunori Ikeguchi (*Grad. Sch. Med. Life Sci., Yokohama City Univ.*)

9:00〜11:30　H 会場（大講義室 A）／Room H (Lecture Room A)

1SHA　新学術領域研究「動的クロマチン構造と機能」共催
生命現象の基本に迫る動的クロマチン構造・機能研究の最前線
Studies of dynamic chromatin structure and function to understand fundamentals of life

オーガナイザー：原口 徳子（(独）情報通信研究機構 未来 ICT 研究所），徳永 万喜洋（東京工業大学）
Organizers: Tokuko Haraguchi (National Institute of Information and Communications Technology), Makio Tokunaga (Tokyo Institute of Technology)

Chromatin, which consists of DNA and proteins, plays a vital role in not only genetic activities but also biological functions. Its structure is not invariable but spatio-temporally varying in response to functions. This dynamic structural change of the chromatin is the very thing that is the basis of the biological functions and activities. Current cutting-edge researches will be presented in wide research fields, structural biology, biophysics, imaging, cell biology and so on. We discuss the basis and meaning of chromatin dynamics.

はじめに
原口 徳子, 徳永 万喜洋
Tokuko Haraguchi, **Makio Tokunaga**

1SHA-01　クロマチンの構造多様性とダイナミクス
Structural versatility and dynamics of chromatin
○胡桃坂 仁志（早稲田大学理工学術院 先進理工学部）
Hitoshi Kurumizaka (*Faculty of Science and Engineering, Waseda University*)

1SHA-02　X 線・中性子溶液散乱によるバリアント・ヌクレオソームの構造研究
Structural Investigation on Variant Nucleosomes by Solution X-ray and Neutron Scattering
○杉山 正明[1], 堀越 直樹[2], 有村 泰宏[2], 井上 倫太郎[1], 越阪部 晃永[2], 胡桃坂 仁志[2]（[1]京大原子炉, [2]早稲田大 電気・情報生命）
Masaaki Sugiyama[1], Naoki Horikoshi[2], Yasuhiro Arimura[2], Rintaro Inoue[1], Akihisa Osakabe[2], Hitoshi Kurumizaka[2] ([1]*KURRI*, [2]*Dept. Elect. Eng. & BioSci., Waseda Univ.*)

1SHA-03　細胞核スケールにおける転写活性分布: ゲノム動力学シミュレーションからのアプローチ
Nuclear-scale spatial distributions of transcriptional activities: an approach from a 3D dynamical simulation model of yeast genome
○徳田 直子, 藤城 新, 笹井 理生（名大院工）
Naoko Tokuda, Shin Fujishiro, Masaki Sasai (*Nagoya Univ.*)

1SHA-04　人工触媒システムによる『合成』エピジェネティクスを目指して
Toward synthetic epigenetics by artificial catalyst systems
○川島 茂裕[1,2], 天本 義史[1,2], 須藤 宏城[1,2], 青井 勇樹[1,2], 永島 臨[1], 越阪部 晃永[3], 有村 泰宏[3], 胡桃坂 仁志[3], 山次 健三[1,2], 金井 求[1,2]（[1]東京大学大学院薬学系研究科, [2]ERATO金井触媒分子生命プロジェクト, [3]早稲田大学先進理工学研究科）
Shigehiro Kawashima[1,2], Yoshifumi Amamoto[1,2], Hiroki Suto[1,2], Yuki Aoi[1,2], Nozomu Nagashima[1], Akihisa Osakabe[3], Yasuhiro Arimura[3], Hitoshi Kurumizaka[3], Kenzo Yamatsugu[1,2], Motomu Kanai[1,2] ([1]*Graduate School of Pharmaceutical Sciences The University of Tokyo*, [2]*ERATO Kanai Life-Science Catalysis Project*, [3]*Graduate School of Advanced Science and Engineering, Waseda University*)

1SHA-05　植物のクロマチン動態メカニズムに迫る
Studies of dynamic chromatin in plants
○松永 幸大[1,2], 平川 健[1], 坂本 勇貴[1], 坂本 卓也[1]（[1]東京理科大学理工学部応用生物科学科, [2]JST, CREST）
Sachihiro Matsunaga[1,2], Takeshi Hirakawa[1], Yuki Sakamoto[1], Takuya Sakamoto[1] ([1]*Dept. Appl. Biol. Sci., Fac. Sci. Tech., Tokyo Univ. Sci.*, [2]*JST, CREST*)

1SHA-06　クロマチン修飾と転写活性化の in vivo ダイナミクス
Chromatin modification and transcription activation in vivo
○木村 宏（東京工業大学大学院生命理工学研究科）
Hiroshi Kimura (*Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology*)

9:00～11:30　I会場（大講義室B）／Room I (Lecture Room B)

1SIA　自然史に学ぶバイオミメティクスの未来
Future of biomimetics learning from natural history

オーガナイザー：飯野 亮太（自然科学研究機構 岡崎統合バイオサイエンスセンター），出口 茂（海洋研究開発機構）
Organizers: Ryota Iino (Okazaki Institute for Integrative Bioscience, National Institutes of Natural Sciences), Shigeru Deguchi (Japan Agency for Marine-Earth Science and Technology)

Biomimetics learns from sophisticated functions and structures of living things, and attracts many chemists and physicist not only as an important theme of basic science but also as a key for innovation to achieve sustainable society. However, chemists and physicists still do not know many, peculiar living things. In this symposium, we invite natural historians studying such interesting living things and researchers of biomimetics at the levels of molecules, organs and individuals. We will try to merge them and discuss the possibility of new biomimetics.

1SIA-01　バイオミメティクス：生物多様性による技術革新
Biomimetics: Innovation Based on Biodiversity
○下村 政嗣（千歳科学技術大学）
Masatsugu Shimomura (*Chitose Institute of Science and Technology*)

1SIA-02　昆虫のふしぎなつくり―驚異の構造と機能
A review of surprising structures and functions of insects
○野村 周平（国立科博・動物）
Shuhei Nomura (*Dept. Zool., Nat. Mus. Nat. Sci., Tokyo*)

1SIA-03　海綿動物に学ぶバイオミメティクスの可能性
Sponges as potential model for biomimetics
○椿 玲未（海洋研究開発機構）
Remi Tsubaki (*JAMSTEC*)

1SIA-04　生物の構造色とその光学効果
Structural color of animals and its optical effects
○吉岡 伸也（東京理科大学）
Shinya Yoshioka (*Tokyo Univ. of Sci.*)

1SIA-05　ヤモリ模擬粘着剤の力学
Mechanics of Gecko Inspired Adhesives
○山口 哲生, 赤峯 彰, 澤江 義則（九大・院工機械）
Tetsuo Yamaguchi, Akira Akamine, Yoshinori Sawae (*Dept. Mech. Eng., Kyushu Univ.*)

1SIA-06　生物に見られる微細・複合構造の生み出す材料物性：濡れ性と破壊靭性の観点から
Wetting and toughness of fine and composite structures in biological materials
○奥村 剛（お茶の水女子大学）
Ko Okumura (*Ochanomizu Univ.*)

9:00～11:30　J会場（105教室）／Room J (Room 105)

1SJA　アクチン線維と結合タンパク質の構造と機能とゆらぎ
Advances in our understanding of the interaction between actin filaments and their binding proteins

オーガナイザー：辰巳 仁史（金沢工業大学），本多 元（長岡技術科学大学）
Organizers: Hitoshi Tatsumi (Kanazawa Institute of Technology), Hajime Honda (Nagaoka University of Technology)

Actin is an abundant, highly conserved protein that polymerizes into filaments. The polymerization and depolymerization of actin filaments is vital for the morphogenesis of dynamic cytoskeletal arrays and actin dependent cell motility. Owing to the importance in biology, the interaction between actin and actin binding proteins has been studied in considerable detail with different methods including single molecule imaging techniques. Recent progresses in this field will be summarized in this symposium.

1SJA-01 アクチン線維のゆらぎはコフィリンのアクチン結合を制御する
Fluctuations of actin filaments modulate the binding of cofilin to the filament
○早川 公英[1], 曽我部 正博[1], 辰巳 仁史[2]（[1]名大院・医・メカノバイオロジー, [2]金沢工大・応用バイオ）
Kimihide Hayakawa[1], Masahiro Sokabe[1], Hitoshi Tatsumi[2] ([1]*Mechanobiology, Nagoya Univ.*, [2]*Dept. of Appl. Biosci., Kanazawa Inst. of Tech.*)

1SJA-02 アクチンフィラメントの協同的構造変化は、アクチン結合タンパク質との相互作用をアロステリックに制御する
Cooperative conformational changes of actin filaments allosterically regulate interactions with actin binding proteins
○上田 太郎[1], 古寺 哲幸[2], 徳楽 清孝[3]（[1]産総研・バイオメディカル, [2]金沢大・バイオAFM, [3]室蘭工大・化）
Taro Uyeda[1], Noriyuki Kodera[2], Kiyotaka Tokuraku[3] ([1]*Biomed. Res. Inst., AIST*, [2]*Bio-AFM FRC, Kanazawa U.*, [3]*Dept. App. Sci., Muroran Inst. Tech.*)

1SJA-03 中性子準弾性散乱による F-アクチン、ミオシン S1 及び水和水のダイナミクス解析
Dynamics of F-actin, myosin subfragment-1 (S1), and their hydration water studied by quasielastic neutron scattering
○松尾 龍人[1], 荒田 敏昭[2], 小田 俊郎[3], 藤原 悟[1]（[1]原子力機構, [2]阪大, [3]兵庫県立大）
Tatsuhito Matsuo[1], Toshiaki Arata[2], Toshiro Oda[3], Satoru Fujiwara[1] ([1]*JAEA*, [2]*Osaka Univ.*, [3]*Univ. Hyogo*)

1SJA-04 アクチン繊維の「状態」とアクチン結合タンパク質
States of an actin filament associated with binding of other proteins
○本多 元[1], 森 一貴[1], 小林 健司[1], 前里 咲良[1], 門間 康平[1], 伊藤 洋貴[1], 石川 良樹[2]（[1]長岡技術科学大学生物専攻, [2]群馬県立県民健康科学大学）
Hajime Honda[1], Kazutaka Mori[1], Kenji Kobayashi[1], Sakura Maesato[1], Kohei Monma[1], Hirotaka Itou[1], Ryoki Ishikawa[2] ([1]*Nagaoka Univ. Tech.*, [2]*Gunma Pref. Col. Health Sci.*)

9:00～11:30　K 会場（107 教室）／Room K (Room 107)

1SKA　膜を介したプロトン透過機構
Proton permeation mechanism across membrane

オーガナイザー：中川 敦史（大阪大学），岡村 康司（大阪大学）
Organizers: Atsushi Nakagawa (Osaka University), Yasushi Okamura (Osaka University)

Proton acts in many important biological processes, such as energy generation and motion etc. In addition to them, it has recently been revealed that proton acts in much more various biological processes, such as regulation of reactive oxygen species and regulation sperm motility, upon pH changes and potential changes across the membrane.
This symposium will discuss the mechanism of proton permeation across the membrane using various techniques, such as computational sciences, spectroscopy and biophysical and biochemical methods, based on the atomic structures of the membrane protein molecules that work in proton permeation.

1SKA-01 V-ATPase のイオン透過機構
Ion transporting mechanism of V-ATPase
○村田 武士[1,2]（[1]千葉大・理, [2]JST・さきがけ）
Takeshi Murata[1,2] ([1]*Science/Chiba-U*, [2]*PRESTO/JST*)

1SKA-02 電位依存性プロトンチャネルを介したプロトン透過経路の構造学的知見
Structural insight into proton conduction pathway via voltage-gated proton channel
○竹下 浩平[1,2,3], 坂田 宗平[4], 山下 栄樹[1], 藤原 祐一郎[4], 岡村 康司[4,5], 中川 敦史[1,5]（[1]阪大蛋白研, [2]阪大未来戦略, [3]JSTさきがけ, [4]阪大院医, [5]JST-CREST）
Kohei Takeshita[1,2,3], Souhei Sakata[4], Eiki Yamashita[1], Yuichiro Fujiwara[4], Yasushi Okamura[4,5], Atsushi Nakagawa[1,5] ([1]*Inst. Protein Res., Osaka Univ.*, [2]*Inst. Acad. Initiat., Osaka Univ.*, [3]*JST-PRESTO*, [4]*Grad. Sch. Med., Osaka Univ.*, [5]*JST-CREST*)

1SKA-03 バクテリアのべん毛運動におけるイオン透過とエネルギー変換
Ion flax and energy transduction in bacterial flagellar motilty
○本間 道夫（名古屋大 院理 生命理学）
Michio Homma (*Nagoya Univ.*)

1SKA-04 プロトンポンプは何を運んでいるのか？
What do H^+ pumps transport?
○神取 秀樹（名古屋工業大学）
Hideki Kandori (*Nagoya Inst. Tech.*)

1SKA-05 分子動力学シミュレーションを用いた電位依存性プロトンチャネル VSOP における亜鉛イオンの影響の解析
Molecular dynamics simulations of the effect of zinc on the voltage-gated proton channel VSOP
○近藤 寛子[1,2,3], 城田 松之[3,4,5], 米澤 康滋[2,6], 鷹野 優[1,2], 木下 賢吾[2,3,5,7]（[1]広島市大・院・情報, [2]JST-CREST, [3]東北大・院・情報, [4]東北大・院・医, [5]東北大・メガバンク, [6]近大・先端研, [7]東北大・加齢研）
Hiroko X. Kondo[1,2,3], Matsuyuki Shirota[3,4,5], Yasushige Yonezawa[2,6], Yu Takano[1,2], Kengo Kinoshita[2,3,5,7] ([1]*GSIS, Hiroshima City Univ.*, [2]*JST-CREST*, [3]*GSIS, Tohoku Univ.*, [4]*Grad. Sch. Med, Tohoku Univ.*, [5]*ToMMo, Tohoku Univ.*, [6]*IAT, Kinki Univ.*, [7]*IDAC, Tohoku Univ.*)

1SKA-06 チトクロム c 酸化酵素の酸化還元反応と共役したプロトン取り込み・排出機構
Molecular mechanism of redox coupled proton uptake and release processes of Cytochrome c oxidase
○重田 育照[1], 神谷 克政[2]（[1]筑波大学大学院数理物質科学研究科, [2]神奈川工科大学基礎・教養教育センター物理）
Yasuteru Shigeta[1], Katsumasa Kamiya[2] ([1]*Department of Pure and Applied Sciences, University of Tsukuba*, [2]*Center for Basic Education and Integrated Learning, Kanagawa Institute of Technology*)

13:20〜16:20　A 会場（大会議室）／Room A (Conference Room)

1SAP　新学術領域研究「温度を基軸とした生命現象の統合的理解」共催
温度生物学の幕開け
The dawn of thermal biology

オーガナイザー：岡部 弘基（東京大学），久原 篤（甲南大学）
Organizers: Kohki Okabe (The University of Tokyo), Atsushi Kuhara (Konan University)

Temperature, a key regulator of biochemical reactions, influences physiological functions such as the metabolism and biological rhythm of organisms. Recent vigorous development in the methodology of intracellular thermometry has urged a novel field of biology focused on temperature, "thermal biology", to emerge. This symposium will overview the latest progress in thermal biology focusing on both cells and organisms, and explore how this fundamental physical parameter contributes to all molecular-based biology.

1SAP-01 生細胞内における温度計測と操作による温度生物学
Imaging and manipulation of intracellular temperature for thermal biology
○岡部 弘基[1,2], 船津 高志[1]（[1]東大院薬, [2]JST さきがけ）
Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

1SAP-02 ASK1 signaling regulates brown adipocyte maturation
Kazuki Hattori, Hidenori Ichijo (*Laboratory of Cell Signaling, Graduate School of Pharmaceutical Sciences, The University of Tokyo*)

1SAP-03 TRP チャネルによる温度感知の分子機構
Moleculae Mechanisms of Thermosensation through TRP channels
○富永 真琴[1,2]（[1]自然科学研究機構岡崎統合バイオサイエンスセンター, [2]総合研究大学院大学）
Makoto Tominaga[1,2] ([1]*Okazaki Institute for Integrative Bioscience*, [2]*SOKENDAI*)

1SAP-04 体温の中枢神経調節
Central regulation of body temperature
○中村 和弘[1,2]（[1]名古屋大・医・統合生理, [2]さきがけ・JST）
Kazuhiro Nakamura[1,2] ([1]*Dept. Integrative Physiol., Nagoya Univ. Grad. Sch. Med.*, [2]*PRESTO, JST*)

1SAP-05 熱ストレスで駆動する核—細胞質間輸送を担う Hikeshi
Thermal stress-induced nucleocytoplasmic transport mediated by Hikeshi
○今本 尚子（理研）
Naoko Imamoto (*RIKEN*)

1SAP-06 局所熱パルス法による単一細胞の機能制御
Regulation of cellular functions by microscopic heat pulses
○大山 廣太郎[1,2], 鈴木 団[3,4], 石渡 信一[2,3,4]（[1]東京慈恵会医科大・細胞生理, [2]早稲田大・先進理工, [3]早稲田バイオサイエンスシンガポール研（WABIOS）, [4]早稲田大・重点機構）
Kotaro Oyama[1,2], Madoka Suzuki[3,4], Shin'ichi Ishiwata[2,3,4] ([1]*Dept. Cell Physiol., The Jikei Univ. Sch. Med.*, [2]*Sch. Adv. Sci. Eng., Waseda Univ.*, [3]*WASEDA Biosci. Res. Inst. Singapore (WABIOS)*, [4]*Org. Univ. Res. Initiatives, Waseda Univ.*)

1SAP-07 線虫 C. elegans における経験に依存した低温馴化機構
Temperature experience-dependent cold acclimation in nematode C. elegans
宇治澤 知代, 大西 康平, 三浦 徹, 太田 茜, ○久原 篤（甲南大学 統合ニューロバイオロジー研究所）
Tomoyo Ujisawa, Kohei Ohnishi, Tohru Miura, Akane Ohta, **Atsushi Kuhara** (*Institute for Integrative Neurobiology, Konan Univ.*)

13:20〜16:20　B 会場（ファカルティホール）／Room B (Faculty Hall)

1SBP　蛋白質・核酸共存系に対する実験家と理論家の挑戦
The challenge of experimentalists and theorists for the protein and nucleic acid system

オーガナイザー：鎌形 清人（東北大学），亀田 倫史（産業技術総合研究所）
Organizers: Kiyoto Kamagata (Tohoku University), Tomoshi Kameda (AIST)

DNA and proteins work cooperatively to maintain functions of a living cell. Transcription is regulated by proteins called as transcription factors, and translation is performed by ribosome, a protein-RNA complex. However, it is difficult to examine concomitant systems of protein and DNA, because methods for investigating them has been limited. In this symposium, experimentalists and theorists using single-molecule measurements, molecular design, MD simulations, and bioinformatics will introduce hot topics in the systems of protein and nucleic acids.

1SBP-01 Target search process of a tumor suppressor p53 revealed by single-molecule fluorescence microscopy
Kiyoto Kamagata (*IMRAM, Tohoku Univ.*)

1SBP-02 分子シミュレーションによる蛋白質-DNA 相互作用の構造可塑性
Flexibility in protein-DNA interactions studied by molecular simulations
○高田 彰二（京大理・生物物理）
Shoji Takada (*Dept. Biophys., Kyoto Univ.*)

1SBP-03 細胞機能の制御に向けた DNA 結合タンパク質の創製
Design of Artificial DNA Binding Proteins towards Manipulation of Cellular Functions
○今西 未来（京大化研）
Miki Imanishi (*ICR, Kyoto Univ.*)

1SBP-04 RNA 結合タンパク質のＲＮＡ界面予測方法の向上とその適用
Improvement in the prediction of RNA interface in RNA-binding Protein
○由良 敬[1,2]（[1]お茶大 生命情報, [2]遺伝研）
Kei Yura[1,2] ([1]*Center for Info. Biol., Ochanomizu Univ.*, [2]*NIG*)

1SBP-05 蛋白質・RNA 複合体の立体構造予測
Three dimensional protein-RNA complex structure prediction
○亀田 倫史[1], 岩切 淳一[2], 浜田 道昭[3], 由良 敬[4,5], 浅井 潔[1,2]（[1]（独）産業技術総合研究所 創薬基盤研究部門, [2]東京大・新領域, [3]早稲田大・理工, [4]お茶の水大・, [5]遺伝研）
Tomoshi Kameda[1], Junichi Iwakiri[2], Michiaki Hamada[3], Kei Yura[4,5], Kiyoshi Asai[1,2] ([1]*Biotech. Research Inst. for Drug Discovery, AIST*, [2]*Grad. School of Frontier Sciences, Univ. of Tokyo*, [3]*Faculty of Science and Engineering, Waseda Univ.*, [4]*Grad. School of Humanities and Sciences, Univ. of Ochanomizu*, [5]*NIG*)

1SBP-06 リボアート・システムを用いたアプタマー創薬
Therapeutics Aptamer Discovery by RiboART System
○中村 義一（東京大学医科学研究所RNA医科学社会連携研究部門）
Yoshikazu Nakamura (*UTokyo IMSUT RNA Medical Science*)

13:20〜16:20　F 会場（AV 講義室）／Room F (AV Lecture Room)

1SFP　膜タンパク質と膜脂質が織りなす協奏と合奏
Concerto and ensemble of membrane proteins and membrane lipids

オーガナイザー：山下 敦子（岡山大学），宮澤 淳夫（兵庫県立大学）
Organizers: Atsuko Yamashita (Okayama University), Atsuo Miyazawa (University of Hyogo)

Biomembrane is a place where phospholipid bilayer and membrane proteins act together for cellular functions. Nevertheless, traditional researches for lipids and membrane proteins have been carried out independently, or we have been prone to listen their functions just as “solo”. In this symposium, we would discuss the structure, function, and dynamics of membrane proteins and membrane lipids side-by-side, and try to listen their “harmony”, so as to understand the biomembrane functions in depth from the viewpoint of their cooperation.

1SFP-01　タンパク質と脂質分子の協調
Cooperation among protein and lipid molecules
○藤吉 好則（名大・院創薬/CeSPI）
Yoshinori Fujiyoshi (*Grad. Sch. Pharma. Sci./CeSPI, Univ. Nagoya*)

1SFP-02　生体膜におけるリン脂質分子の運動と機能
Probing the molecular motion and function of membrane phospholipids
○梅田 真郷（京大・工 合成・生物化学専攻）
Masato Umeda (*Dept. Syth.Chem and Biol.Chem. Sch. Eng. Kyoto Univ.*)

1SFP-03　Dynamics of Pore Forming Toxins during Their Assembling on Lipid Membranes
Neval Yilmaz, Toshihide Kobayashi (*RIKEN*)

1SFP-04　Structure and Dynamics of Membrane Lipid Nanodomains using High-Speed AFM
Pierre-Emmanuel Milhiet (*CBS, CNRS/INSERM*)

1SFP-05　High-Speed Atomic Force Microscopy: Watching Dynamic Processes at the Membrane at High Spatio-Temporal Resolution
Lorena Redondo, Atsushi Miyagi, Ignacio Casuso, **Simon Scheuring** (*INSERM/Aix-Marseille U.*)

1SFP-06　脂質分子が膜タンパク質機能に及ぼす役割
Role of lipid molecules on molecular functions of membrane proteins
○杉田 有治（独立行政法人 理化学研究所・杉田理論分子科学研究室）
Yuji Sugita (*RIKEN Theoretical Molecular Science Laboratory*)

13:20〜16:20　G 会場（レクチャーホール）／Room G (Lecture Hall)

1SGP　X 線自由電子レーザー施設 SACLA が拓く生物物理研究の新展開
New developments in biophysical studies with X-ray free electron laser at the SACLA

オーガナイザー：城地 保昌（高輝度光科学研究センター），湯本 史明（高エネルギー加速器研究機構）
Organizers: Yasumasa Joti (JASRI/SPring-8), Fumiaki Yumoto (KEK)

X-ray free electron lasers (XFELs) provide intense and coherent X-rays with ultra-short pulse duration. A “diffraction-before-destruction” concept was proposed as a new experimental scheme using XFELs. The concept has been demonstrated at XFEL facilities including SACLA, Japanese XFEL facility, for protein crystals and non-crystalline biological particles. In this symposium, we will introduce recent outcomes in applications to biological systems at the SACLA. Furthermore, new developments toward biophysical studies in future will be discussed.

1SGP-01　SACLA の現状と生物学研究実験
Current Status of SACLA and Experiments for Biological Studies
○登野 健介[1,2]（[1]高輝度研, [2]理研・放射光セ）
Kensuke Tono[1,2] ([1]*JASRI*, [2]*RIKEN SPring-8 Center*)

1SGP-02 オルガネラの XFEL 低温コヒーレント X 線回折イメージング
Cryogenic coherent X-ray diffraction imaging of cellular organelle by using XFEL
関口 優希[1,2], 小林 周[1,2], 苣口 友隆[1,2], ○中迫 雅由[1,2], 高山 裕貴[2], 山本 雅貴[2], 乾 弥生[3], 松永 幸大[3], 市川 雄一[4], 胡桃坂 仁志[4], 清水 光弘[5]（[1]慶應大学・理工, [2]理研・RSC, [3]東京理科大・理工, [4]早稲田大・理工, [5]明星大・理工）
Yuki Sekiguchi[1,2], Amane Kobayashi[1,2], Tomotaka Oroguchi[1,2], **Masayoshi Nakasako**[1,2], Yuki Takayama[2], Masaki Yamamoto[2], Yayoi Inui[3], Sachihiro Matsunaga[3], Yuichi Ichikawa[4], Hitoshi Kurumizaka[4], Mitsuhiro Shimizu[5] ([1]*Sci. Tech., Keio Univ*, [2]*RIKEN SPring-8 Center*, [3]*Sci. Tech., Tokyo Univ. Sci.*, [4]*Sci. Tech., Waseda Univ.*, [5]*Sci. Tech., Meisei Univ.*)

1SGP-03 X 線レーザー回折による生細胞イメージング
Imaging live cells by X-ray laser diffraction
○西野 吉則[1], 木村 隆志[1], 城地 保昌[2], 別所 義隆[3]（[1]北大・電子研, [2]（公財）高輝度光科学研究センター, [3]IoP, Academia Sinica）
Yoshinori Nishino[1], Takashi Kimura[1], Yasumasa Joti[2], Yoshitaka Bessho[3] ([1]*RIES, Hokkaido Univ.*, [2]*JASRI/SPring-8*, [3]*IoP, Academia Sinica*)

1SGP-04 重原子誘導体を利用したシリアルフェムト秒結晶学による新規構造決定
De novo structure determination for serial femtosecond crystallography using heavy atom derivatives
○山下 恵太郎[1], 潘 東青[2], 菅原 道泰[1], 吾郷 日出夫[1], 山本 雅貴[1], 中津 亨[1,2]（[1]理研SPring-8, [2]京大院薬・構造生物）
Keitaro Yamashita[1], Dongqing Pan[2], Michihiro Sugahara[1], Hideo Ago[1], Masaki Yamamoto[1], Toru Nakatsu[1,2] ([1]*RIKEN/SPring-8*, [2]*Dep. Struct. Biol., Grad. Sch. Pharm. Sci., Kyoto Univ.*)

1SGP-05 SACLA における時分割シリアルフェムト秒結晶構造解析：バクテリオロドプシンの光反応中間体構造について
Structural dynamics of bacteriorhodopsin using time-resolved serial femtosecond crystallography at SACLA
○南後 恵理子（理化学研究所 放射光科学総合研究センター）
Eriko Nango (*RIKEN RSC*)

1SGP-06 巨大タンパク質の高分解能・無損傷結晶構造解析が可能なフェムト秒 X 線結晶構造解析法の開発
Development of a method of femtosecond crystallography enabling determination of high-resolution native structure of huge proteins
○吾郷 日出夫[1], 平田 邦生[1,2], 上野 剛[1], 山本 雅貴[1], 伊藤-新澤 恭子[3], 月原 冨武[2,3,4], 吉川 信也[3], 菅 倫寛[5], 秋田 総理[5], 沈 建仁[5]（[1]理研・放射光科学総合研究センター, [2]科学技術振興機構・CREST, [3]兵県大・院生命理学, [4]阪大・蛋白研, [5]岡大・院自然科学）
Hideo Ago[1], Kunio Hirata[1,2], Go Ueno[1], Masaki Yamamoto[1], Kyoko Shinzawa-Itoh[3], Tomitake Tsukihara[2,3,4], Shinya Yoshikawa[3], Michihiro Suga[5], Fusamichi Akita[5], Jian-Ren Shen[5] ([1]*RIKEN SPring-8 Center*, [2]*CREST, JST*, [3]*Grad. Sch. Sci., Univ. Hyogo*, [4]*Institute for Protein Research, Osaka Univ.*, [5]*Grad. Sch. Nat. Sci., Okayama Univ.*)

13:20〜16:20　H 会場（大講義室 A）／Room H (Lecture Room A)

1SHP　チューブリンの構造スイッチングとその生理的意義
Conformational switching of tubulin and its physiological function

オーガナイザー：上村 慎治（中央大学）, 武藤 悦子（理化学研究所）
Organizers: Shinji Kamimura (Chuo University), Etsuko Mutoh (RIKEN)

Microtubules play a variety of functions in cells. Recent studies using high-resolution cry-electron micrography and X-ray fiber diffraction revealed that multiple conformational states of tubulin give rise to structural polymorphism in microtubule lattice, which may be crucial for diverse physiological functions of microtubules. Multiple conformations of tubulin also underlie dynamic instability of microtubules, where the balance between assembly and disassembly is stochastically switched in a nucleotide-dependent manner. In this session, we introduce the forefront in the field of microtubule, aiming to share new findings and insights about the conformational dynamics of tubulin and microtubule.

1SHP-01 X 線繊維回折法による微小管内チューブリン構造の動態解析
Microtubule dynamics revealed by the X-ray fiber diffraction analysis
○上村 慎治[1], 岩本 裕之[2]（[1]中央大学・理工学部・生命科学科, [2]JASRI, SPring-8）
Shinji Kamimura[1], Hiroyuki Iwamoto[2] ([1]*Dept. Biol. Sci., Chuo Univ.*, [2]*JASRI, SPring-8*)

1SHP-02 The structure of kinesin bound to tubulin links the nucleotide cycle to movement
Benoit Gigant (*I2BC, CNRS, France*)

1SHP-03 微小管における構造柔軟性と、モーター蛋白の運動メカニズム
Conformational flexibility of microtubule linked to motility mechanism of motor proteins
○武藤 悦子（理化学研究所 脳科学総合研究センター）
Etsuko Muto (*RIKEN, Brain Science Institute*)

1SHP-04 Conformational switching of tubulin serves as the guidance cue for the intracellular transport by kinesin
Yasushi Okada (*Quantitative Biology Center, RIKEN*)

1SHP-05 Conformational switching of tubulin by Alp7/14 TOG-family polymerase
Frauke Hussmann, Douglas R Drummond, Daniel Peet, Douglas S Martin, **Robert A Cross** (*Warwick Medical School*)

1SHP-06 ダイナミックな微小管細胞骨格は植物細胞の形を作り出し、環境ストレスに応答する
The dynamic microtubule cytoskeleton builds the shape of plant cells and responds to environmental stresses
○橋本 隆（奈良先端科学技術大学院大学バイオサイエンス研究科）
Takashi Hashimoto (*Graduate School of Biological Sciences, Nara Institute of Science and Technology*)

13:20〜16:20　I会場（大講義室B）／Room I (Lecture Room B)

1SIP　超解像光学顕微鏡のライフサイエンスへの展開
Principles and applications of super resolution microscopy in recent life science

オーガナイザー：加藤 薫（産業技術総合研究所），原田 慶恵（京都大学）
Organizers: Kaoru Katoh (AIST), Yoshie Harada (Kyoto University)

Super resolution microscopy reveals fine structures smaller than resolution limit of conventional optical microscopes. This symposium deals with principles and applications of typical super resolution microscopy (SIM, STED, Airy Scan and localization microscopes (STORM, PALM, etc.)) available for life sciences. We will demonstrate a variety of present biological applications, including cytoskeleton, chromosomes, intracellular organella and plant cells. Moreover, we will also discuss limitation and possibility of future applications.

1SIP-01 アクチン系細胞骨格の SIM 及び STED による観察
Observation of actin-based cytoskeletal structures with SIM and STED microscopy
○加藤 薫[1], 上条 桂樹[2]（[1]産総研・バイオメディカル, [2]東北大・医・人体構造）
Kaoru Katoh[1], Keijyu Kamijo[2] ([1]*Biomed. Res. Inst., AIST*, [2]*Dept. Anat. & Anthropol., Tohoku Univ. Sch. of Med.*)

1SIP-02 構造化照明顕微鏡ＳＩＭの原理
Principle of the structured illumination microscopy
○照井 勇輝（ニコン）
Yuki Terui (*Nikon*)

1SIP-03 リアルタイム超解像イメージングに向けた高速多重蛍光分子アルゴリズム Wedged Template Matching（WTM）の開発
Development of super resolution multi-emitter algorithm using template matching; towards real time analysis and visualization
竹嶋 智親[1], 高橋 輝夫[1], 山下 慈郎[1], 岡田 康志[2], ○渡部 重夫[1]（[1]浜松ホトニクス株式会社 システム事業部, [2]理研QBiC）
Tomochika Takeshima[1], Teruo Takahashi[1], Jiro Yamashita[1], Yasushi Okada[2], **Shigeo Watanabe**[1] ([1]*Hamamatsu Photonics K.K.*, [2]*RIKEN QBiC*)

1SIP-04 ピンホールを超えた共焦点超解像顕微鏡システム Airyscan
Airyscan: a new confocal based superresolution microscopy
○渡邉 俊之[1], ヴァイスハルト クラウス[2]（[1]カールツァイスマイクロスコピー, [2]カールツァイスマイクロスコピー）
Toshiyuki Watanabe[1], Klaus Weisshart[2] ([1]*Carl Zeiss Microscopy*, [2]*Carl Zeiss Microscopy GmbH*)

1SIP-05 超解像蛍光顕微鏡法が明らかにするストレス顆粒内内在性 mRNA のナノスケール空間構成
Super-resolution fluorescence microscopy reveals nanoscale spatial organization of endogenous mRNA in stress granules
○菅原 皓（東大・院薬）
Ko Sugawara (*Grad. Sch. Pharm. Sci., Univ. of Tokyo*)

1SIP-06 超解像イメージングで解き明かす植物細胞膜切断装置のダイナミクス
Directional assembly of plant endocytic dynamin unveiled by super-resolution imaging
○藤本 優[1], 中野 明彦[2,3], 上田 貴志[2,4]（[1]東大院・農学生命科学, [2]東大院・理, [3]理研・光量子工学, [4]JST・さきがけ）
Masaru Fujimoto[1], Akihiko Nakano[2,3], Takashi Ueda[2,4] ([1]*Grad. Sch. Agric. Life Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*RIKEN Center for Advanced Photonics*, [4]*PRESTO, JST*)

1SIP-07 Imaging local sphingomyelin domains in the plasma membrane using lipid-specific probes and super-resolution microscopy
Toshihide Kobayashi, Mitsuhiro Abe (*RIKEN*)

1SIP-08 Meiotic chromosome structure and function visualized with super-resolution microscopy
Peter Carlton (*iCeMS, Kyoto Univ.*)

第 2 日目（9 月 14 日（月））／Day 2（Sep. 14 Mon.）

8:45〜11:15　A 会場（大会議室）／Room A (Conference Room)

2SAA　新学術領域研究「動的構造生命科学が拓く新発想測定技術」共催
動的構造生命科学を拓く新発想測定技術
Novel measurement techniques for visualizing 'live' protein molecules at work

オーガナイザー：西田 紀貴（東京大学），神田 大輔（九州大学）
Organizers: Noritaka Nishida (The University of Tokyo), Daisuke Kohda (Kyushu University)

Static, but accurate 3D protein structures determined by X-ray crystallography have provided convenient explanations of the protein functions as biological functional elements. However, we still need to understand the more details of the dynamic aspects of protein structures in transient states under physiological conditions. Our Grant-in-Aid for Scientific Research on Innovative Areas aims to upgrade NMR, AFM, and crystallographic techniques dramatically as innovative measurement techniques for protein dynamics. In this symposium, we will introduce several new measurement techniques, and discuss their applicabilities and limitations.

2SAA-01 タンパク質結晶内に創りだした隙間を利用して、タンパク質分子内部の運動分布を解析する
Crystal contact-free space for analyzing spatial distribution of protein internal motions
○神田 大輔（九大・生医研・構造生物）
Daisuke Kohda (*Struct. Biol., Med. Inst. Bioreg., Kyushu Univ.*)

2SAA-02 蛋白質の分子揺らぎと構造変化を計測する X 線 1 分子動態計測法の開発
Refinements of the Diffracted X-ray Tracking Method for Recording Molecular Fluctuations and Conformational Changes of Proteins
○清水 啓史（福井大 医 分子生理）
Hirofumi Shimizu (*Univ. Fukui. Fac. Med. Sci.*)

2SAA-03 細胞内クラウディング環境における蛋白質のフォールディングとダイナミクスを NMR で観察する
NMR approaches to investigate protein folding and dynamics in the crowded intracellular environment
○伊藤 隆[1,2]（[1]首都大, [2]CREST/JST）
Yutaka Ito[1,2] ([1]*Tokyo Metropolitan Univ.*, [2]*CREST/JST*)

2SAA-04 Cytoplasmic conformational transition of Sec translocon
Yoshiki Tanaka[1], Yasunori Sugano[1], Mizuki Takemoto[2], Takaharu Mori[3], Takamitsu Haruyama[4], Arata Furukawa[1], Tsukasa Kusakizako[2], Kaoru Kumazaki[2], Ayako Kashima[2], Ryuichiro Ishitani[2], Hiroki Konno[4], Yuji Sugita[3], Osamu Nureki[2], **Tomoya Tsukazaki**[1,5] ([1]*NAIST*, [2]*Grad. Sch. of Sci., Univ. of Tokyo*, [3]*RIKEN*, [4]*BioAFM-FRC, Kanazawa Univ.*, [5]*JST, PRESTO*)

2SAA-05 大規模構造変化を伴う反応モデル構築のためのマルチレゾリューションシミュレーション手法の開発
Development of multi-resolution simulation methods for reactions with large conformational changes in biological system
○小林 千草[1], 松永 康佑[1], Jung Jaewoon[1,2], 杉田 有治[1,2,3,4]（[1]理研、AICS, [2]理研、杉田理論分子科学, [3]理研、iTHES, [4]理研、QBiC）
Chigusa Kobayashi[1], Yasuhiro Matsunaga[1], Jaewoon Jung[1,2], Yuji Sugita[1,2,3,4] ([1]*RIKEN, AICS*, [2]*RIKEN, TMS*, [3]*RIKEN, iTHES*, [4]*RIKEN, QBiC*)

2SAA-06 The identification of the ankyrin repeat domain as a novel lipid-binding module
Shiro Suetsugu[1], Nobuaki Takahashi[2], Yuzuru Itoh[3], Kazuhiro Takemura[3], Akio Kitao[3], Yasuo Mori[2] ([1]*Graduate School of Biological Sciences, Nara Institute of Science and Technology*, [2]*Graduate School of Engineering, Kyoto University*, [3]*Institute of Molecular and Cellular Biosciences, University of Tokyo*)

8:45～11:15　D 会場（103 教室）／Room D (Room 103)

2SDA　脂質活性構造研究の最前線
Frontier of the study of lipid-active structures

オーガナイザー：杉山 成（大阪大学），前仲 勝実（北海道大学）
Organizers: Shigeru Sugiyama (Osaka University), Katsumi Maenaka (Hokkaido University)

Lipids are involved in crucial biological functions, including the components of biomembrane, signaling, and storing energy. To understand these functions in molecular level, it is important to elucidate the structures and dynamics of lipid molecules, interacting with protein molecules. However, lipid molecules show conformational flexibility and interact with protein molecules in non-specific manner. These physic-chemical properties of lipid are very bottleneck in studies on lipid active structures. In this symposium, we will present some recent studies on lipid active structures.

Open discussion

2SDA-01 膜蛋白質－膜脂質の相互作用解析を目指した重元素化脂質の利用
Application of synthetic lipids for analysis of interaction between membrane protein and lipids
○杉山 成[1,2]（[1]阪大院・理, [2]JST, ERATO）
Shigeru Sugiyama[1,2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*JST, ERATO*)

2SDA-02 脂質ラフトの NMR 解析
NMR Studies of Lipid Rafts
○松森 信明[1], 村田 道雄[2,3]（[1]九大・院理, [2]阪大・院理, [3]ERATO）
Nobuaki Matsumori[1], Michio Murata[2,3] ([1]*Grad. Sch. Sci., Kyushu Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*ERATO*)

2SDA-03 FABP3 脂肪酸結合における内部水分子の重要性
Interplay between Internal Water Molecules and Bound Fatty Acid Molecule in FABP3 Binding Cavity
○松岳 大輔[1,2], 松岡 茂[1,2], 杉山 成[1,2], 村田 道雄[1,2]（[1]阪大院理, [2]ERATO村田プロジェクト）
Daisuke Matsuoka[1,2], Shigeru Matsuoka[1,2], Shigeru Sugiyama[1,2], Michio Murata[1,2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*ERATO Murata Project*)

2SDA-04 Structural basis for glycolipid recognition of human immune receptor, Mincle, and rational inhibitor design
Atsushi Furukawa[1], Takanori Matsumaru[2], Risa Ikeno[1], Yusuke Shuchi[1], Sho Yamasaki[3], Toyoyuki Ose[1], **Katsumi Maenaka**[1,2] ([1]*Laboratory of Biomolecular Science, Faculty of Pharmaceutical Sciences, Hokkaido University*, [2]*Center for Research and Education on Drug Discovery, Faculty of Pharmaceutical Sciences, Hokkaido University*, [3]*Division of Molecular Immunology, Research Center for Infectious Diseases, Medical Institute of Bioregulation, Kyushu University*)

2SDA-05 新規部分フッ素化リン脂質膜の創製
Development of Novel Partially Fluorinated Phospholipid Membrane for Biophysical Studies of Membrane Proteins
○園山 正史（群馬大・院理工）
Masashi Sonoyama (*Div. Mol. Sci., Fac. Sci. & Tech., Gunma Univ.*)

2SDA-06 膜孔形成毒素の自己組織化に生体膜の組成とダイナミクスが与える影響の分子基盤
Molecular basis of self-assembly of a transmembrane hemolytic toxin triggered by specific membrane composition and dynamics
田中 耕路[1], Caaveiro Jose[1], ○津本 浩平[1,2]（[1]東京大学大学院　工学系研究科, [2]東京大学　医科学研究所）
Koji Tanaka[1], Jose Caaveiro[1], **Kouhei Tsumoto**[1,2] ([1]*School of Engineering, The University of Tokyo*, [2]*The Institute of Medical Science, The University of Tokyo*)

8:45～11:15　F 会場（AV 講義室）／Room F (AV Lecture Room)
2SFA　大自由度ダイナミクスから“生きている状態”の記述へ
Description of 'living state' based on high-dimensional cellular dynamics

オーガナイザー：古澤 力（理化学研究所），金子 邦彦（東京大学）
Organizers: Chikara Furusawa (RIKEN), Kunihiko Kaneko (The University of Tokyo)

Recent advances in technologies provide huge amounts of quantitative data, and one major challenge here is to extract the essential (macroscopic) variables from such high-dimensional data to understand the dynamic behavior of systems, including development, adaptation, and evolution. In this symposium, we will discuss both experimental and theoretical analysis based on such high-dimensional data for better understanding of complex biological systems.

2SFA-01 動物胚が発生と進化の法則性、砂時計モデルに従うのはなぜか？
What makes animal embryos to follow the hourglass model?
○入江 直樹（東大・院理）
Naoki Irie (*Grad. Sch. Sci., Univ. Tokyo*)

2SFA-02 腸内細菌叢由来代謝産物がもたらす生体恒常性維持機構
Gut microbiota-derived metabolites shape host physiological homeostasis
○福田 真嗣（慶大・先端生命研）
Shinji Fukuda (*Inst. Adv. Biosci., Keio Univ.*)

2SFA-03 シングルセル遺伝子発現解析が明らかにする細胞分化のメカニズム
Single Cell Transcriptome Analysis Dissects Cell Fate Specification
○渡辺 亮（京都大学）
Akira Watanabe (*Kyoto University*)

2SFA-04 Dynamics of phenotype-genotype mapping in laboratory evolution of Escherichia coli
Chikara Furusawa (*QBiC, RIKEN*)

2SFA-05 進化と適応における揺らぎ、応答の普遍関係
Fluctuation and Response in Adaptation and Evolution-Universal Relationship
○金子 邦彦（東大総合文化）
Kunihiko Kaneko (*Univ. of Tokyo, Departmet of Basic Science*)

8:45～11:15　G 会場（レクチャーホール）／Room G (Lecture Hall)
2SGA　新学術領域研究「理論と実験の協奏による柔らかな分子系の機能の科学」共催
生体分子の機能を制御する柔らかさ
Functions of biomolecules controlled by flexibility

オーガナイザー：神取 秀樹（名古屋工業大学），林 重彦（京都大学）
Organizers: Hideki Kandori (Nagoya Institute of Tecnology), Shigehiko Hayashi (Kyoto University)

Biomolecules are typical soft molecular systems, which possess many degrees of freedom and show enhanced functional performance, due to their ability to flexibly change their structures. The ultimate goal in the study is to understand how biomolecules are controlled by softness to create function. In this symposium, speakers present recent findings on their own soft molecular systems using theory, advanced spectroscopy, and biomolecular engineering. Role of softness in creation of function will be discussed.

2SGA-01 高速 AFM で明らかにする分子シャペロン ClpB ーの柔らかさと機能
Structural Flexibility and Chaperone Activity of ClpB observed by High-Speed AFM
○内橋 貴之[1,2,3], 渡辺 洋平[4], 飯野 亮太[5], 安藤 敏夫[1,2,3]（[1]金沢大理工, [2]金沢大バイオAFM-FRC, [3]CREST-JST, [4]甲南大理工, [5]岡崎統合バイオ/分子研）
Takayuki Uchihashi[1,2,3], Yo-hei Watanabe[4], Ryota Iino[5], Toshio Ando[1,2,3] ([1]*Coll. Sci. & Eng. Kanazawa Univ.*, [2]*Bio-AFM FRC, Kanazawa Univ.*, [3]*CREST-JST*, [4]*Dep. Biol., Konan Univ.*, [5]*OIIB/IMS*)

2SGA-02 Theoretical study of ion transport pathway of channelrhodopsin
Norio Yoshida (*Kyushu Univ.*)

2SGA-03 固体 NMR によるファラオニスフォボロドプシンの Tyr174 と Tyr199 の水素結合変化の解析
Solid-state NMR study of hydrogen-bonding alterations of Tyr174 and Tyr199 in pharaonis phoborhodopsin
○川村 出（横浜国立大学大学院工学府）
Izuru Kawamura (*Grad. Sch. Eng., Yokohama Natl. Univ.*)

2SGA-04 カリウムイオンを感知して活性がスイッチングするインテリジェントリボザイム/アプタマーの創製
Development of intelligent ribozyme/aptamer that sense K^+ and switch on their activities
○片平 正人（京大エネ理工）
Masato Katahira (*Inst. Adv. Energ., Kyoto Univ.*)

2SGA-05 蛋白質分子の機能的運動を制御するナノスケールの水和構造変化
Nanoscale wetting and drying processes dominate protein functional motions
○荏口 友隆, 中迫 雅由（慶應義塾大学・物理学科）
Tomotaka Oroguchi, Masayoshi Nakasako (*Sci. Tech., Keio Univ.*)

2SGA-06 立体選択的な水酸化酵素をめざしたマンガンポルフィセン錯体を含むミオグロビンの設計と構築
Design and engineering of myoglobin containing a manganese porphycene toward enantioselective hydroxylase
○大洞 光司（阪大院工）
Koji Ohora (*Grad. Sch. Eng. Osaka Univ.*)

8:45～11:15　H 会場（大講義室 A）／Room H (Lecture Room A)

2SHA　新学術領域研究「新生鎖の生物学」共催
新生鎖の合成と構造形成過程に潜む生物物理学
Biophysics underlying the synthesis and folding of nascent polypeptide chains

オーガナイザー：稲葉 謙次（東北大学），田中 元雅（理化学研究所）
Organizers: Kenji Inaba (Tohoku University), Motomasa Tanaka (RIKEN)

For the past several years, an increasing number of studies have demonstrated that nascent polypeptide chains that are still on the way of translation interact with several factors including molecular chaperones, ubiquitin ligases and endonucleases to ensure the protein and messenger RNA quality control in cells. However, mechanisms of operations of these cellular systems remain to be elucidated. In this symposium, speakers will show their latest experimental data produced by advanced biophysical approaches and discuss molecular basis underlying the synthesis and folding of nascent polypeptide chains.

2SHA-01 Single-molecule analysis of conformational space of a yeast prion protein Sup35NM using optical tweezers
Yusuke Komi[1], Rodrigo Maillard[2], Piere Rodriguez[2], Carlos Bustamante[2,3], Motomasa Tanaka[1] ([1]*BSI, RIKEN*, [2]*UC Berkeley*, [3]*HHMI*)

2SHA-02 タンパク質翻訳の 1 分子研究
Single molecule study of protein translation
○上村 想太郎（東京大学大学院理学系研究科）
Sotaro Uemura (*Graduate School of Science, The University of Tokyo*)

2SHA-03 SecM による翻訳停止の分子メカニズムに関する研究
Study on the molecular mechanism of translation arrest by SecM
楊 倬皓, 飯塚 怜, 郭 遠芳, ○船津 高志（東大・院薬）
Zhuohao Yang, Ryo Iizuka, Yuanfang Guo, **Takashi Funatsu** (*Grad. Sch. Phar. Sci., Univ. Tokyo*)

2SHA-04 Incorporation of fluorescent amino acid for lifetime FRET
Takanori Uzawa (*RIKEN*)

2SHA-05 シャペロニン空洞内フォールディングにおける疎水性相互作用の役割
The role of hydrophobic tethering in chaperonin-mediated folding
○元島 史尋[1], 吉田 賢右[2]（[1]富山県立大・工・生物工学セ, [2]京都産業大学）
Fumihiro Motojima[1], Masasuke Yoshida[2] ([1]*Biotech. Res. Cent. and Dept. of Biotech., Toyama Pref. Univ.*, [2]*Kyoto Sangyo Univ.*)

2SHA-06 高速 AFM が明らかにする PDI の酸化的フォールディングの触媒機構の解明
A new mechanism of operation of PDI during its catalysis of oxidative protein folding revealed by high-speed atomic force microscopy
○奥村 正樹[1], 野井 健太郎[2,5], 金村 進吾[1], 増井 翔史[1], 引間 孝明[3], 秋山 修志[3,4], 小椋 光[2,5], 稲葉 謙次[1,5]（[1]東北大学・多元研, [2]熊大・発生研, [3]理研播磨, [4]分子研, [5]CREST, JST）
Masaki Okumura[1], Kentaro Noi[2,5], Shingo Kanemura[1], Shoji Masui[1], Takaaki Hikima[3], Shuji Akiyama[3,4], Teru Ogura[2,5], Kenji Inaba[1,5] ([1]*IMRAM, Tohoku University*, [2]*IMEG, Kumamoto University*, [3]*RIKEN Harima*, [4]*IMS*, [5]*CREST, JST*)

2SHA-07 小胞体トランスロコンによる新生鎖のハンドリング
Handling of nascent chain by ER translocon during membrane protein integration
○阪口 雅郎（兵県大・院生命理学）
Masao Sakaguchi (*Grad. Sch. Life Sci., Univ. Hyogo*)

8:45～11:15　I 会場（大講義室 B）／Room I (Lecture Room B)

2SIA　新学術領域研究「生命分子システムにおける動的秩序形成と高次機能発現」共催
ATP 加水分解が介在する時空間動秩序の形成
Formation of spatiotemporal dynamic ordering mediated by ATP hydrolysis

オーガナイザー：内山 進（大阪大学），杉山 正明（京都大学）
Organizers: Susumu Uchiyama (Osaka University), Masaaki Sugiyama (Kyoto University)

Living systems are characterized as dynamic processes of assembly and disassembly of various biomolecules that are self-organized, interacting with the external environment.
In this symposium, we will focus on the living systems where dynamic structural change and assembly/disassembly occurs continuously according to ATP hydrolysis state, such as protein clock systems, molecular motor, and actin assembly/disassembly processes. Recent findings related to the spatiotemporal dynamic ordering in these systems will be introduced.

はじめに
内山 進
Susumu Uchiyama

2SIA-01 ３つのタンパク質で再構成できるシアノバクテリアの生物時計
A self-sustaining and temperature-compensated circadian rhythm reconstituted *in vitro* in a minimal system containing three Kai proteins
○寺内 一姫（立命館大生命科学）
Kazuki Terauchi (*Dep. Life Sci., Ritsumeikan Univ.*)

2SIA-02 KaiC as Circadian Pacemaker of Cyanobacterial Circadian Clock
Shuji Akiyama (*Institute for Molecular Science*)

2SIA-03 KaiABC システムにおける ATP 加水分解と概日振動の結合についてのメゾスケールモデリング
Mesoscale modelling of coupling between ATP hydrolysis and circadian oscillation in KaiABC system
Das Sumita, 橋本 翔太, 寺田 智樹, ○笹井 理生（名古屋大学大学院工学研究科計算理工学専攻）
Sumita Das, Shota Hashimoto, Tomoki P. Terada, **Masaki Sasai** (*Department of Computational Science and Engineering, Nagoya University*)

2SIA-04 ATP 駆動分子モーターの 1 分子高速可視化解析
Single-molecule high-speed imaging analysis of ATP-driven molecular motors
○飯野 亮太（自然科学研究機構 岡崎統合バイオ・分子研）
Ryota Iino (*OIIB and IMS, NINS*)

2SIA-05 細胞の力覚応答と増殖におけるアクチン動的秩序の役割
Roles of actin dynamic ordering in mechanosensing and cell proliferation
○水野 健作（東北大・生命科学）
Kensaku Mizuno (*Grad. Sch. Life Sci., Tohoku Univ.*)

2SIA-06 生体溶液中での ATP 加水分解による同符号電荷間引力制御と時空間動秩序
Spatiotemporal dynamic ordering regulated by ATP hydrolysis and effective attraction between negatively charged sites in a biofluid
○秋山 良（九州大院理化学）
Ryo Akiyama (*Dept. of Chem., Kyushu Univ.*)

おわりに
杉山 正明
Masaaki Sugiyama

8:45～11:15　J 会場（105 教室）／Room J (Room 105)

2SJA　物質と生命の境界を探る合成生物学
Synthetic biology exploring the border between life and material

オーガナイザー：市橋 伯一（大阪大学），津留 三良（大阪大学）
Organizers: Norikazu Ichihashi (Osaka University), Saburo Tsuru (Osaka University)

Molecular biology has revealed various sophisticated mechanisms of the cell. However, we still do not know the principle that allows the emergence of life from an assembly of molecules. In this symposium, we invited prominent researchers exploring the Interdisciplinary field between life science and material science. They are reconstituting artificial cell-life systems in a test tube or massively engineering the natural cell. We would like to discuss the significance and the future of this new field of science.

2SJA-01 人工細胞の進化的動態
Evolutionary dynamics of an artificial cell model
○四方 哲也[1,2]（[1]大阪大学大学院情報科学研究科バイオ情報工学専攻, [2]大阪大学大学院生命機能研究科）
Tetsuya Yomo[1,2] ([1]*Grad. Sch. Info. Sci. & Tech., Osaka Univ.*, [2]*Grad. Sch. Front. Bio-Sci., Osaka Univ.*)

2SJA-02 二成分脂質ベシクルで探る生命と分子集合体の境界
A border between cellular life and molecular assembly revealed by binary lipid vesicles
○佐久間 由香（東北大・院理学）
Yuka Sakuma (*Grad. Sch. Sci., Univ. Tohoku*)

2SJA-03 バクテリアサイボーグ
Bacterial cyborg; integrated bacterial cell systems into arrayed lipid bilayer chamber
○野地 博行（東大工学研究科）
Hiroyuki Noji (*School of Engineering, The University of Tokyo*)

2SJA-04 ゲノム合成生物学でのゲノム構築
Scrapping and Building Bacteria Genomes for Novel Synthetic Genomics
○板谷 光泰（慶應大学・先端生命研）
Mitsuhiro Itaya (*Inst. Adv. Biosce. Keio Univ.*)

2SJA-05 Mutation accumulation of bacterial genome toward genomic inactivation
Saburo Tsuru, Atsushi Shibai (*Osaka Univ.*)

8:45〜11:15　K 会場（107 教室）／Room K (Room 107)

2SKA　GPCR の多様性、そしてその機能理解へ至る多様なアプローチ
Diversity of GPCRs, and multimodal approach to understanding their functions

オーガナイザー：佐藤 恵太（京都大学），柳川 正隆（理化学研究所）
Organizers: Keita Sato (Kyoto University), Masataka Yanagawa (RIKEN)

G protein-coupled receptors (GPCRs) are involved in various physiological and pathological processes including vision, taste, olfaction, and endocrine systems. This is due to the diversification and adaptation of GPCRs during the course of molecular evolution. In this symposium, young scientists will talk about how diversified GPCRs work, and how to elucidate the molecular machinery and the physiological function of GPCRs using multimodal methodologies such as NMR, molecular simulation, single molecule measurement, and cell biological techniques. We would like to discuss future aspects of research on GPCRs.

2SKA-01　Opn5L1 は光サイクル性の反応で制御される G タンパク質共役型受容体である
Opn5L1 is a photocyclic GPCR
○佐藤 恵太[1], 山下 高廣[1], 大内 淑代[2], 竹内 敦子[3], 友成 さゆり[4], 酒井 佳寿美[1], 今元 泰[1], 和田 昭盛[3], 七田 芳則[1]（[1]京大・院理, [2]岡山大・院医歯薬, [3]神戸薬大, [4]徳島大・ソシオテクノサイエンス）
Keita Sato[1], Takahiro Yamashita[1], Hideyo Ohuchi[2], Atsuko Takeuchi[3], Sayuri Tomonari[4], Kazumi Sakai[1], Yasushi Imamoto[1], Akimori Wada[3], Yoshinori Shichida[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*, [3]*Kobe Pharm. Univ.*, [4]*Inst. Tech. Sci., Univ. Tokushima*)

2SKA-02　Evolution of the umami taste receptor in vertebrates
Yasuka Toda[1,2] ([1]*Kikkoman Corp.*, [2]*Dept. Appl. Biol. Chem., The Univ. Tokyo*)

2SKA-03　膜型 TGFα 切断を利用した GPCR 活性化の簡便な検出法とその応用
TGFα shedding serves a useful readout for dissecting GPCR signaling
○井上 飛鳥[1,2]（[1]東北大・薬, [2]JSTさきがけ）
Asuka Inoue[1,2] ([1]*Grad. Sch. of Pharm. Sci., Tohoku Univ.*, [2]*PRESTO, JST*)

2SKA-04　NMR 法を用いた脂質二重膜中の GPCR の動的構造解析
Functional dynamics of G-protein-coupled receptors in lipid bilayers revealed by NMR
○幸福 裕[1], 上田 卓見[1,2], 奥出 順也[1], 白石 勇太朗[1], 近藤 啓太[1], 水村 拓也[1], 鈴木 志歩[1], 嶋田 一夫[1]（[1]東大・院薬系, [2]JST・さきがけ）
Yutaka Kofuku[1], Takumi Ueda[1,2], Junya Okude[1], Yutaro Shiraishi[1], Keita Kondo[1], Takuya Mizumura[1], Shiho Suzuki[1], Ichio Shimada[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*PRESTO, JST*)

2SKA-05　バイオインフォマティクスによる GPCR オリゴマー研究
Bioinformatics approaches in the study of GPCR oligomers
○根本 航[1], 山西 芳裕[2], Limviphuvadh Vachiranee[3], 齊藤 哲[1], 藤代 峻輔[1], 雨宮 雄一[1], 藤 博幸[4]（[1]東京電機大・理工・生命理工, [2]九大・生防医, [3]BII, A*STAR, SINGAPORE, [4]関西学院大・理工学部・生命医化）
Wataru Nemoto[1], Yoshihiro Yamanishi[2], Vachiranee Limviphuvadh[3], Akira Saito[1], Shunsuke Fujishiro[1], Yuichi Amemiya[1], Hiroyuki Toh[4] ([1]*Div. of Life Sci. & Eng., TDU, Japan*, [2]*MiB, Kyushu Univ., Japan*, [3]*BII, A*STAR, Singapore*, [4]*Dept. of Biomed. Chem., Sch. of Sci. & Tech., Kwansei Gakuin Univ., Japan*)

2SKA-06　1 分子イメージングで見る GPCR の多量体化とエンドサイトーシス
Single-molecule imaging of GPCR oligomerization followed by internalization
○柳川 正隆（理研）
Masataka Yanagawa (*RIKEN*)

13:55〜16:25　A 会場（大会議室）／Room A (Conference Room)

2SAP　新学術領域研究「運動超分子マシナリーが織りなす調和と多様性」共催
生体マシナリーにおける力発生と進化の共通原理
Principles and evolution for force generation in bio-nanomachines

オーガナイザー：南野 徹（大阪大学），宮田 真人（大阪市立大学）
Organizers: Tohru Minamino (Osaka Universiy), Makoto Miyata (Osaka City University)

Recent advancements on single molecule manipulation and imaging technologies combined with high-resolution structural analysis allow scientists to understand the force generation mechanism at the atomic levels in various systems. Now, we can deeply think beyond the framework and the data, to discuss about the principles and evolution for force generation in bio-nanomachinery. In this symposium, nine speakers will introduce their own and related systems briefly, and claim for the common principles.

Intorduction
宮田 真人
Makoto Miyata

2SAP-01　熱揺らぎは分子モーターに共通する駆動力なのだろうか？
Is thermal fluctuation a common driving force of molecular motors?
○中村 修一（東北大・院工）
Shuichi Nakamura (*Grad. Sch. Eng., Tohoku Univ.*)

2SAP-02　F_1-ATPase における異種エネルギー変換機構
Heterogeneous energy conversion mechanism of F_1-ATPase
○渡邉 力也[1,2]（[1]東京大学大学院工学系研究科応用化学専攻, [2]JST・さきがけ）
Rikiya Watanabe[1,2] ([1]*Department of Applied Chemistry, The University of Tokyo*, [2]*PRESTO, JST*)

2SAP-03　高速 AFM により導かれたミオシン V の力発生の原理
Principle for force generation in myosin V illustrated by high-speed AFM
○古寺 哲幸[1,2], 内橋 貴之[1,3,4], 安藤 敏夫[1,3,4]（[1]金沢大・理工・バイオAFM, [2]JST/さきがけ, [3]金沢大・理工・数物科学, [4]JST/CREST）
Noriyuki Kodera[1,2], Takayuki Uchihashi[1,3,4], Toshio Ando[1,3,4] ([1]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [2]*PRESTO, JST*, [3]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [4]*CREST, JST*)

2SAP-04　分子機械動作機構のクーロン的視点
Coulombic viewpoint of how molecular machines work
○高野 光則（早大 先進理工 物理）
Mitsunori Takano (*Dept. Phys., Waseda Univ.*)

2SAP-05　タンパク質分泌を駆動する反復モーターの作動原理
The working principle of repetitive motors driving protein export
○森 博幸（京大・ウイルス研）
Hiroyuki Mori (*Inst. Virus Res., Kyoto Univ.*)

2SAP-06　RNA ポリメラーゼの力発生機構
Force generation mechanism of the RNA polymerase
○原田 慶恵（京都大学 iCeMS）
Yoshie Harada (*iCeMS, Kyoto University*)

2SAP-07　１分子から階層を超えて細胞そして組織の機能へ繋ぐ
Bridging the hierarchy of single molecule and cellular and tissue functions
○柳田 敏雄[1,2]（[1]理研QBiC, NiCT, [2]阪大　CiNet）
Toshio Yanagida[1,2] ([1]*RIKEN QBiC; NiCT*, [2]*Osaka Univ. CiNet*)

2SAP-08 疑おう
Let's doubt ourselves
○木下 一彦（早大理工・物理）
Kazuhiko Kinosita, Jr. (*Dept. Phys., Fac. Sci. Eng., Waseda Univ.*)

2SAP-09 F1-ATPase が 100%近いエネルギー効率で回転する仕組み
A mechanism how F1-ATPase rotates with near 100% energy efficiency
○吉田 賢右, 秡田 英一郎（Kyoto Sangyo Univ.）
Masasuke Yoshida, Ei-ichiro Saita (*Kyoto Sangyo Univ.*)

Open discussion

13:55〜16:25　F 会場（AV 講義室）／Room F (AV Lecture Room)

2SFP　ナノとマクロを繋ぐ生物電子顕微鏡アプローチ
Advanced electron microscopy: A new world view of mesoscale biology

オーガナイザー：安永 卓生（九州工業大学），太田 啓介（久留米大学）
Organizers: Takuo Yasunaga (Kyushu Institute of Technology), Keisuke Ohta (Kurume University)

Morphological understanding of meso-scale structures or organisms, such as cells and organelle, is essential for elucidation of their higher order functions of organisms from the macroscopic views. It is, conventionally, difficult to observe the three-dimensional structure at the nano-scale levels but advanced electron microscopy (EM) such as electron tomography and serial block-face SEM, makes it possible to observe them at the molecular level. Here, we introduce the recent progress of EM in cooperation with Japanese Society of Microscopy.

2SFP-01 ナノとマクロを繋ぐ生物電子顕微鏡アプローチ
Advanced electron microscopy: A new world view of mesoscale biology
○安永 卓生（九工大・情報工・生命情報工）
Takuo Yasunaga (*Dept. of Biosci. and Bioinfo., Sch. of Comp. Sci. and Sys. Eng., Kyushu Inst. Tech.*)

2SFP-02 クライオ電子線トモグラフィは細胞内タンパク質複合体構造解析に有力な手法である
Cryo-electron tomography is one of powerful techniques to understand the structures of intracellular protein complexes
○荒牧 慎二[1], 真柳 浩太[2], 青山 一弘[3,4], 安永 卓生[3,4]（[1]九工大・情報工・生命情報工学, [2]九大・防医研, [3]日本エフイー・アイ株式会社, [4]阪大・超高圧電子顕微鏡センター）
Shinji Aramaki[1], Kouta Mayanagi[2], Kazuhiro Aoyama[3,4], Takuo Yasunaga[3,4] ([1]*Dept. of Bioscience and Bioinformatics, Kyushu Inst. of Tech.*, [2]*Medical Inst. of Bioregulation Kyushu Univ.*, [3]*FEI Company*, [4]*Research Center for Ultra-High Voltage EM, Osaka Univ.*)

2SFP-03 電子顕微鏡によって明らかになったシヌクレインタンパク質の分子特性から細胞内輸送における役割を探る
Molecular properties of synuclein found through the electron microscopy and its function in the microtubule-based intracellular transport
○鳥羽 栞（大阪市立大学医学部 細胞機能制御学教室（第2生化））
Shiori Toba (*Graduate School of Medicine, Osaka City University*)

2SFP-04 低温電子線トモグラフィーにおける電子線直接検知型カメラの効果
The power of electron direct detector for electron cryotomography
○川本 晃大[1], 森本 雄祐[1,2], 加藤 貴之[1], 難波 啓一[1,2]（[1]阪大院・生命機能, [2]理研・QBiC）
Akihiro Kawamoto[1], V.Yusuke Morimoto[1,2], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ*, [2]*QBiC, RIKEN*)

2SFP-05 Serial block-face SEM による細胞分裂の方向を決定する新規細胞膜構造の観察
A novel plasma membrane structure to determine the orientation of the centrosome during cell division revealed by serial block-face SEM
○宮崎 直幸[1], 根岸 剛文[2], 上野 直人[2], 村田 和義[1]（[1]生理研, [2]基生研）
Naoyuki Miyazaki[1], Takefumi Negishi[2], Naoto Ueno[2], Kazuyoshi Murata[1] ([1]*NIPS*, [2]*NIBB*)

2SFP-06 SEM ベース三次元再構築法（FIB-SEM トモグラフィー法）を用いたミトコンドリア-小胞体の相互関係の可視化
3D organization of the mitochondria-associate membrane in mammalian cells by using FIB-SEM tomography
○太田 啓介[1,2]（[1]久留米大学医学部解剖学講座, [2]理研・生命システム研究センター）
Keisuke Ohta[1,2] ([1]*Dept. Anatomy, Kurume Univ. Sch. Med.*, [2]*RIKEN, Qbic*)

13:55〜16:25　G 会場（レクチャーホール）／Room G (Lecture Hall)

2SGP　蛋白質の相互作用と動的立体構造変化
Dynamical structural change of proteins upon interaction with biomolecules

オーガナイザー：星野 大（京都大学），原田 英里砂（サントリー生命科学財団）
Organizers: Masaru Hoshino (Kyoto University), Erisa Harada (Suntory Foundation for Life Sciences)

Recent advances in protein science revealed that a protein structure is not necessarily a unique “static” conformation, but is changing dynamically depending on the environment. Such a variety of conformational change is closely related to the protein functions, exemplified by a coupled folding and binding of intrinsically disordered proteins, and fibril formation of amyloidogenic proteins. In this symposium, we would like to shed light on the “dynamic” nature of proteins upon interaction with biomolecules by using a various biophysical techniques.

2SGP-01 Sox2 の DNA 結合ドメインの構造揺らぎから分かった DNA 認識機構
DNA binding mechanism of high-mobility group box domain of sox2 revealed by its conformational flexibility
○原田 英里砂[1], 小沼 剛[2], 森 祥子[1], 菅瀬 謙治[3]（[1]（公財）サントリー生命科学財団, [2]マウントサイナイ医科大学, [3]京都大学大学院）
Erisa Harada[1], Tsuyoshi Konuma[2], Shoko Mori[1], Kenji Sugase[3] ([1]*Suntory Foundation for Life Sciences*, [2]*Mount Sinai School of Medicine*, [3]*Kyoto University*)

2SGP-02 Dynamic Recognition of Unfolded Proteins by the Trigger Factor Chaperone as Investigated by NMR
Tomohide Saio[1], Xiao Guan[2], Paolo Rossi[2], Charalampos Kalodimos[2] ([1]*Div. of Chem., Grad. School of Sci., Hokkaido Univ.*, [2]*CIPR, Rutgers University*)

2SGP-03 Structural characterization of a denaturant denatured state
Hironari Kamikubo (*MS, NAIST*)

2SGP-04 天然物の結合によるヒトトランスサイレチンの構造変化
Structural changes upon the binding of natural products in human transthyretin
○横山 武司（富山大・薬）
Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

2SGP-05 Protein-Protein interactions of the apicoplast proteins of Plasmodium falciparum
Takashi Saitoh[1], Shohei Yuasa[2], Fumina Oosaka[1], Katsumi Maenaka[1], Toshiharu Hase[2], Yoko Kimata-Ariga[2] ([1]*Grad. Sch. of Pharma. Sci., Hokkaido Univ.*, [2]*Inst. for Protein Research, Osaka Univ.*)

2SGP-06 リガンドとの相互作用に伴う膜蛋白質の動的立体構造変化
Dynamical structural change of membrane proteins upon interaction with their ligands
○上田 卓見[1,2], 幸福 裕[1], 嶋田 一夫[1]（[1]東大院薬, [2]さきがけ）
Takumi Ueda[1,2], Yutaka Kofuku[1], Ichio Shimada[1] ([1]*Grad. Sch. Pharm. Sci. the Univ. of Tokyo*, [2]*PRESTO, JST*)

13:55〜16:25　H 会場（大講義室 A）／Room H (Lecture Room A)

2SHP　ポンプ、酵素、モーター、機能の鍵：pKa
Key role of pKa on functions of pump, enzyme, and motor

オーガナイザー：吉田 紀生（九州大学），田中 伊知朗（茨城大学）
Organizers: Norio Yoshida (Kyushu University), Ichiro Tanaka (Ibaraki University)

Protonation of ionizable amino acids plays a key role in characterizing the protein functions. Spectroscopic studies have clearly indicated the importance of protonation regarding light-driven proton pumps. Recently, discussions in pKa of amino acids have been extended to various functions of protein, such as channel, motor, and enzyme, the situation of which requires further studies both in experiments and theories. In this symposium, importance of pKa in protein functions will be discussed from a general standpoint of methods and targets.

オープニング
吉田 紀生
Norio Yoshida

2SHP-01　プロトネーション状態の確認だけでなく機能探索の手法としての生体高分子中性子結晶学
Neutron protein crystallography as the technique for not only the identification of protonation state but also function investigation
○田中 伊知朗[1,2]（[1]茨城大工, [2]茨城大フロンティア）
Ichiro Tanaka[1,2] ([1]*Coll. of Eng., Ibaraki Univ.*, [2]*Frontier Res. Center*)

2SHP-02　生体分子水和水の構造とカイネティクスの分子動力学解析
Atomic-Scale View of Biomolecular Hydration: From Structure to Kinetics
○米谷 佳晃（日本原子力研究開発機構 量子ビーム応用研究センター）
Yoshiteru Yonetani (*Japan Atomic Energy Agency, Quantum Beam Science Center*)

2SHP-03　タンパク質機能における内部結合水の役割
Role of bound water molecules inside proteins in their functional processes
○林 重彦（京都大学大学院理学研究科化学専攻）
Shigehiko Hayashi (*Grad. Sch. Sci., Kyoto Univ.*)

2SHP-04　膜チャネル・トランスポーターの分子機構
Molecular Mechanisms of Membrane Channel/Transporter
○濡木 理（東京大学大学院理学系研究科生物化学専攻）
Osamu Nureki (*Department of Biological Science, Graduate School of Science, The University of Tokyo*)

2SHP-05　ナトリウムポンプ型ロドプシンの機能におけるプロトンの役割
The role of proton on the function of sodium pump rhodopsin
○井上 圭一（名古屋工業大学）
Keiichi Inoue (*Nagoya Institute of Technology*)

2SHP-06　チトクロム酸化酵素の高分解能 X 線結晶構造解析から明らかとなった、酸素還元反応と共役したプロトン輸送機序
High-resolution X-ray structural analysis reveals how cytochrome c oxidase pumps protons coupled with molecular oxygen reduction
○島田 敦広[1], 矢野 直峰[2], 岸田 佳織[3], 馬場 淳平[1], 江藤 勇樹[1], 波多野 啓太[1], 山下 栄樹[4], 伊藤-新澤 恭子[1], 月原 冨武[1,4], 吉川 信也[1]（[1]兵県大・生命理・ピコバイオロジー研, [2]茨城大・フロンティア応用原子研, [3]神大・院農・生命機能・応用生命, [4]阪大・蛋白研）
Atsuhiro Shimada[1], Naomine Yano[2], Kaori Kishida[3], Junpei Baba[1], Yuki Etoh[1], Keita Hatano[1], Eiki Yamashita[4], Kyoko Shinzawa-Itoh[1], Tomitake Tsukihara[1,4], Shinya Yoshikawa[1] ([1]*Picobiol. Inst., Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Front. Res. Cent. Appl. Atomic Sci., Ibaraki Univ.*, [3]*Appl. Chem. in Biosci., Agrobio., Grad. Sch. Agri. Sci. Kobe Univ.*, [4]*Inst. Protein Res., Osaka Univ.*)

2SHP-07　Brownian ratchet mechanisms of macromolecular motors
Keiichi Namba (*Graduate School of Frontier Biosciences, Osaka University*)

クロージング
田中 伊知朗
Ichiro Tanaka

13:55～16:25　I 会場（大講義室 B）／Room I (Lecture Room B)

2SIP　高次の生命現象を可能にする類似構造による多様なリガンド認識機構
Mechanisms of diverged ligand recognition by similar protein structures for higher-order biological processes

オーガナイザー：深井 周也（東京大学），大戸 梅治（東京大学）
Organizers: Shuya Fukai (The University of Tokyo), Umeharu Ohto (The University of Tokyo)

In higher-order biological processes such as immunity and neurodevelopment, receptors recognize a wide variety of ligands that substantially differ in their size and shape. On the other hand, structures of such receptors are composed of limited sets of 'basic' folds such as leucine-rich repeat and immunoglobulin-like fold. In this symposium, we will discuss mechanisms by which membrane receptors of similar structures can recognize a wide variety of ligands, based on recent structural insights into the mechanisms and molecular evolution.

2SIP-01　シナプス分化を制御するスプライスインサート暗号の解読メカニズム
Decoding mechanisms of splice-insert signaling codes for synaptic differentiation
○深井 周也[1,2,3]（[1]東大•放射光•生命科学, [2]東大•分生研, [3]JST CREST）
Shuya Fukai[1,2,3] ([1]*Life Sci. Div., SRRO, Univ. Tokyo*, [2]*IMCB, Univ. Tokyo*, [3]*JST CREST*)

2SIP-02　小脳シナプス形成を担う GluRδ2-Cbln1-neurexin 接着分子複合体の構造基盤
Structural insights into trans-synaptic GluRδ2-Cbln1-neurexin adhesion complex for cerebellar synapse formation
○植村 健[1,2]（[1]信大・学術院・医・分子細胞生理, [2]独立行政法人科学技術振興機構, CREST）
Takeshi Uemura[1,2] ([1]*Dept. Mol. Cell. Physiol., Inst. of Med., Acad. Assy., Shinshu Univ.*, [2]*CREST, JST*)

2SIP-03　Solution behavior of TLR9 studied by analytical ultracentrifugation
Susumu Uchiyama (*Grad. Sch. Eng., Osaka Univ.*)

2SIP-04　細胞表面受容体の結晶解析によって明らかになったシグナル伝達を制御する低親和性相互作用部位
Crystallographic analyses of cell-surface receptors revealed the presence of low-affinity interfaces regulating the signal transduction
○禾 晃和（横浜市立大学大学院生命医科学研究科）
Terukazu Nogi (*Grad. Sch. Med. Lif. Sci., Yokohama City Univ.*)

2SIP-05　グルクロン酸糖結合レクチンとしてのクロトー共受容体
α-klotho is a high affinity lectin that binds terminal glucuronyl residues
○前田 良太[1,2]（[1]コペンハーゲン大, [2]先端医・セ）
Ryota Maeda[1,2] ([1]*Copenhagen Univ.*, [2]*IBRI*)

2SIP-06　Toll 様受容体によるリガンド認識とシグナル伝達
Ligand recognition and signal transduction by Toll-like receptor
○大戸 梅治（東京大学大学院薬学系研究科）
Umeharu Ohto (*Graduate School of Pharmaceutical Sciences, The University of Tokyo*)

9:00〜12:00　A 会場（大会議室）／Room A (Conference Room)

3SAA　JST/CREST「ライフサイエンスの革新を目指した構造生命科学と先端的基盤技術」共催
細胞に近づく構造生命科学の最前線
Frontiers of structural life science approaching to cellular phenomena

オーガナイザー：安藤 敏夫（金沢大学），吉川 雅英（東京大学）
Organizers: Toshio Ando (Kanazawa University), Masahide Kikkawa (The University of Tokyo)

Structural biology has made a big success in solving the structure of proteins that function alone. Nevertheless, it is still a challenge to solve the entire structure of supramolecular ensembles comprising multiple molecules as well as to understand cellular phenomena based on protein structures. In this symposium, we will discuss frontier studies confronting these difficulties to create the next generation of structural life science.

3SAA-01　繊毛・鞭毛の 96 nm 周期を決める分子ものさし
A molecular ruler determines the repeat length in eukaryotic cilia and flagella
○吉川 雅英（東京大学・医・生体構造）
Masahide Kikkawa (*The University of Tokyo*)

3SAA-02　細胞膜へのフォスファチジルセリンの暴露を制御するフリッパーゼとスクランブラーゼ
Flippase and scramblase that regulate the phosphatidylserine-exposure to plasma membrane
○長田 重一（大阪大学免疫学フロンテア研究センター）
Shigekazu Nagata (*Immunology Frontier Research Center, Osaka University*)

3SAA-03　CRISPR-Cas9 の結晶構造
Crystal structure of CRISPR-Cas9
○西増 弘志（東京大学大学院理学系研究科）
Hiroshi Nishimasu (*The University of Tokyo*)

3SAA-04　25 サブユニットからなる転写メディエーター複合体の再構成
Total reconstitution of the 25-subunit Mediator complex of transcription regulation
○今崎 剛[1,2,3], ガーボー パパイ[2,4], 山田 健太郎[2], パトリック シュルツ[4], 高木 雄一郎[2]（[1]JSTさきがけ研究者, [2]インディアナ大, [3]理研, [4]IGBMC）
Tsuyoshi Imasaki[1,2,3], Papi Gabor[2,4], Kentaro Yamada[2], Schultz Patrick[4], Yuichiro Takagi[2] ([1]*JST researcher*, [2]*Indiana Univesity*, [3]*RIKEN*, [4]*IGBMC*)

3SAA-05　オートファジーの始動を担う Atg1/ULK 複合体の構造と機能
Structure and function of the autophagy initiating Atg1/ULK complex
○野田 展生[1,2]（[1](公財)微化研, [2]JST, CREST）
Nobuo N. Noda[1,2] ([1]*Inst. Microbial Chem.*, [2]*JST, CREST*)

9:00〜12:00　B 会場（ファカルティホール）／Room B (Faculty Hall)

3SBA　最新ハイブリッドアプローチによる機能構造解析
State-of-the-art hybrid methods for structural analysis of macromolecular complexes at functional state

オーガナイザー：岩崎 憲治（大阪大学），フロハンス タマ（理化学研究所）
Organizers: Kenji Iwasaki (Osaka University), Florence Tama (RIKEN)

Hybrid methods are nowadays commonly used to analyze higher-order structures of biological molecules. These methods comprise X-ray crystallography, computational methods and lower resolution experimental approaches such as EM, SAXS. Such a multiple approach is essential to interpret the functional state of macromolecular complexes in cellular context. In this symposium, we review these cutting edge methods. Six researchers from the fields of EM, SAXS, and computational methods will introduce their brand new results on this topic.

はじめに
岩崎 憲治
Kenji Iwasaki

3SBA-01 Hybrid Approaches to Characterize Structure and Dynamics of Biomolecular Systems from Single Molecule Experiments
Florence Tama[1,2] ([1]*Nagoya University, Physics*, [2]*RIKEN AICS*)

3SBA-02 2D hybrid analysis: An approach to build 3D atomic model from 2D EM image
Atsushi Matsumoto (*Japan Atomic Energy Agency*)

3SBA-03 分子動力学シミュレーションと電子顕微鏡像を用いたハイブリットシミュレーションによるリボソーム内 tRNA 転位の解析
Analysis of tRNA translocation through the ribosome by a hybrid-simulation using an MD simulation and electron microscopy density maps
○石田 恒（日本原子力研究開発機構量子ビーム応用研究センター分子シミュレーショングループ）
Hisashi Ishida (*Japan Atomic Energy Agency, Quantum Beam Science Center, Molecular Modeling and Simulation Group*)

3SBA-04 X 線溶液散乱と二次構造情報によるタンパク質の立体構造の構築
Construction of Protein Structure by Small-Angle X-ray Scattering Constraints and Secondary Structural Information
○小島 正樹, 森本 康幹, 市岡 隆幸（東京薬科大学）
Masaki Kojima, Yasumasa Morimoto, Takayuki Ichioka (*Tokyo University of Pharmacy and Life Sciences*)

3SBA-05 様々な顕微鏡法によるアクチンフィラメント構造解析
Structural analysis of the actin filament by several microscopy techniques
○成田 哲博[1,2]（[1]名古屋大、理、構造生物学研究センター, [2]JST さきがけ）
Akihiro Narita[1,2] ([1]*Struct. Biol. Res. Center, Nagoya Univ.*, [2]*JST PRESTO*)

9:00～12:00　C 会場（101 教室）／Room C (Room 101)
3SCA　電気生理学的アプローチによる膜タンパク質構造機能相関
Electrophysiological approaches for structure and function of membrane proteins

オーガナイザー：藤原 祐一郎（大阪大学），中條 浩一（大阪医科大学）
Organizers: Yuichiro Fujiwara (Osaka University), Koichi Nakajo (Osaka Medical College)

Electrophysiology has been successfully used to study many aspects of ion channels and transporters including single-molecule behaviors, macroscopic properties and even intermolecular interactions with high time resolution. In combination with diverse approaches (structural biology, molecular imaging, etc.) recently introduced to this research field, electrophysiology is now increasing its importance. The purpose of this symposium is to introduce electrophysiological methods for the general membrane protein researcher. The symposium will cover the theoretical backgrounds and some practical examples of electrophysiological analysis combined with other techniques.

はじめに

3SCA-01 流動電位測定により明らかにされたカリウムチャネルポア内でのイオンと水の共役的透過
The flux coupling between ion and water in the narrow pore of the K^+ channels revealed by the streaming potential measurement
○岩本 真幸, 老木 成稔（福井大・医・分子生理）
Masayuki Iwamoto, Shigetoshi Oiki (*Dept. Mol. Physiol. Biophys., Univ. Fukui Facult. Med. Sci.*)

3SCA-02 電気生理で膜輸送体の ATP 加水分解メカニズムを解明する
Monitoring ATP-hydrolysis cycle by electro-physiological approach: Patch-clamp recordings of CFTR
○相馬 義郎（慶應義塾大・医・薬理）
Yoshiro Sohma (*Keio Univ. Sch. Med.*)

3SCA-03 オメガ-3 脂肪酸は Slo1 BK チャネルを活性化する
Omega-3 fatty acids activate Slo1 BK channels
○田嶋 信義[1], Tian Yutao[2], Xu Rong[2], Heinemann Stefan[3], Hou Shangwei[4], Hoshi Toshinori[2]（[1]金沢医科大学 医学部 生理学Ⅰ, [2]ペンシルベニア大学 生理学, [3]フリードリッヒシラー大学イエナ 生物物理, [4]上海交通大学 システム生物医学センター）
Nobuyoshi Tajima[1], Yutao Tian[2], Rong Xu[2], Stefan Heinemann[3], Shangwei Hou[4], Toshinori Hoshi[2] ([1]*Dep. Physiol., Kanazawa Medical University*, [2]*Dep. Physiol., University of Pennsylvania*, [3]*Center for Molecular Biomedicine, Dep. Biophys., Friedrich Schiller University Jena*, [4]*Shanghai Center for Systems Biomedicine, Shanghai Jiao Tong University*)

3SCA-04 破骨細胞における細胞膜 V-ATPase 電流の経時的解析
Real-time analysis of V-ATPase (proton pump) currents in the plasma membrane of osteoclasts
○久野 みゆき（大阪市立大・院・医・分子細胞生理学）
Miyuki Kuno (*Dept. Physiol., Osaka City Univ. Grad. Sch. Med.*)

3SCA-05 電位依存性 H^+チャネル動的構造への電気生理学的アプローチ
Electrophysiological approaches to structural dynamics of the voltage-gated H^+ channel
○藤原 祐一郎（阪大・院医学）
Yuichiro Fujiwara (*Grad. Sch. of Med., Osaka Univ.*)

3SCA-06 蛍光を使ってイオンチャネルの電流と電位センサーの動きを同時に測定する
Simultaneous recordings of ionic currents and voltage sensor movements by voltage clamp fluorometry
○中條 浩一（大阪医大・医・生理）
Koichi Nakajo (*Dept. Physiol., Osaka Med. Coll.*)

おわりに

9:00～12:00　D 会場（103 教室）／Room D (Room 103)
3SDA　輸送膜タンパク質のダイナミクス
Dynamics of membrane transport proteins

オーガナイザー：鈴木 裕（旭川医科大学），表 弘志（岡山大学）
Organizers: Hiroshi Suzuki (Asahikawa Medical University), Hiroshi Omote (Okayama Unversity)

Ion and phospholipid transport across the membrane plays essential roles in the biological system from beginning of life on the earth. Understanding of molecular mechanisms underlying the pumps and transporters is major subject of current biology. Recent cooperation of biochemical and biophysical studies on the transport system makes this field to enter a new stage. In this session, we will discuss fundamental mechanisms of the transport system as part of energy transduction system.

3SDA-01 胃プロトンポンプのアンタゴニスト結合構造
Antagonist-bound structures of gastric proton pump
○阿部 一啓[1,2]（[1]名大・細胞生理セ, [2]名大院・創薬）
Kazuhiro Abe[1,2] ([1]*CeSPI, Nagoya Univ.*, [2]*Grad. Sch. Pham., Nagoya Univ.*)

3SDA-02 Ca^{2+}-ATPase の触媒部位と輸送部位間の M2 ヘリックスを介したロングレンジ共役
Long-range Coupling between Catalytic and Transport Sites via Second Transmembrane Helix (M2) in Ca^{2+}-ATPase
○大保 貴嗣, 山崎 和生, ダンコ ステファニア, 鈴木 裕（旭川医大・生化）
Takashi Daiho, Kazuo Yamasaki, Stefania Danko, Hiroshi Suzuki (*Biochem., Asahikawa Med. Univ.*)

3SDA-03 Na+,K+-ATPase は膜貫通結合部位に結合した K を順番に置換する- X 線結晶解析を用いたキネティックス測定
Sequential substitution of bound K+ in the transmembrane binding sites of Na+,K+-ATPase, Kinetics by X-ray crystallography
○小川 治夫[1], 平田 絢美[1], Cornelius Flemming[2], 豊島 近[1]（[1]東京大学分子細胞生物学研究所, [2]Department of Biomedicine, Aarhus University）
Haruo Ogawa[1], Ayami Hirata[1], Flemming Cornelius[2], Chikashi Toyoshima[1] ([1]*IMCB, The University of Tokyo*, [2]*Department of Biomedicine, Aarhus University*)

3SDA-04 リン脂質 flippase である P4-ATPase の基質特異性と細胞機能の関係
Phospholipid flippase activities and substrate specificities of P4-ATPases and their roles in cellular function
○申 惠媛（京都大学・院薬学研究科）
Hye-Won Shin (*Grad. Sch. Pharm. Sci., Kyoto Univ.*)

3SDA-05 マラリア原虫のクロロキン耐性トランスポーター(PfCRT)の生成再構成系を用いた機能解析
Functional analysis of Plasmodium falciparum chloroquine resistance transporter (PfCRT) by reconstituted system with purified protein
○表 弘志[1], 樹下 成信[2], 森山 佐和子[1], 宮地 孝明[2], 河上 麻美代[1], 岩井 遥香[1], 福井 智也[1], 森山 芳則[1]（[1]岡山大学大学院医歯薬, [2]岡山大学自然生命支援センター）
Hiroshi Omote[1], Narinobu Juge[2], Sawako Moriyama[1], Takaaki Miyaji[2], Mamiyo Kawakami[1], Haruka Iwai[1], Tomoya Fukui[1], Yoshinori Moriyama[1] ([1]*Dept. of Membrane Biochemistry, Okayama University*, [2]*Adv. Science Research Center, Okayama University*)

3SDA-06 集合に共役した細菌べん毛モーター固定子ユニットの活性化機構
Assembly-coupled activation of the torque-generating stator units in the bacterial flagellar motor
○小嶋 誠司（名古屋大・院理・生命理学）
Seiji Kojima (*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*)

3SDA-07 膜輸送蛋白等の計測における 1 分子顕微鏡観察とマイクロデバイスの活用
Single-molecule observation and utilization of microdevice for measurements of membrane transport proteins
○政池 知子[1,2,3]（[1]東京理科大・理工・応用生物科学, [2]東京理科大・総合研究院, [3]JST・さきがけ）
Tomoko Masaike[1,2,3] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Science*, [2]*Res. Inst. for Sci. and Tech., Tokyo Univ. of Science*, [3]*PRESTO, JST*)

9:00～12:00　F 会場（AV 講義室）／Room F (AV Lecture Room)

3SFA　細胞を診て操作する生物物理的アプローチ
New biophysical approaches to explore and manipulate cells

オーガナイザー：石渡 信一（早稲田大学），馬渕 一誠（学習院大学）
Organizers: Shin'ichi Ishiwata (Waseda University), Issei Mabuchi (Gakushuin University)

We will discuss the development of novel biophysical techniques and experimental systems including both live cells and artificially reconstituted systems for studying the structure and functions of cells. The new techniques and systems are not merely restricted to imaging the localization and dynamics of cellular components under microscope, but are intended to explore the physical states of cells and to manipulate the cellular components such as cell membrane, organelles, and cytoskeletons by externally applying force, heat, and light.

はじめに
石渡 信一
Shin'ichi Ishiwata

3SFA-01 微生物に含まれる脂質の構造解析のための MS-AFM-IR システム
MS-AFM-IR platform for structural analysis of lipid inclusions in micro-organisms
○Lesniewska Eric, Vitry P., Dazzi A., Virolle M−J., Tetard L., Bourillot E.（ICB UMR CNRS 6303, Univ. of Bourgogne Franche-Comte）
Eric Lesniewska, P. Vitry, A. Dazzi, M.−J. Virolle, L. Tetard, E. Bourillot (*ICB UMR CNRS 6303, Univ. of Bourgogne Franche-Comte*)

3SFA-02 原子間力顕微鏡：細胞力学特性の個性を測る
Atomic force microscopy: Quantifying mechanical variation in living cell system
○岡嶋 孝治（北大・情報科学）
Takaharu Okajima (*Grad. Sch. Inform. Sci. Tech., Hokkaido Univ.*)

3SFA-03 新しい熱プローブを用いた細胞発熱計測および分子モーター活性制御
New thermal probes for cellular heat measurement and for temporal regulation of motor proteins
○井上 裕一（東北大・多元研）
Yuichi Inoue (*IMRAM, Tohoku Univ.*)

3SFA-04 細胞構造と機能を操る高圧力顕微鏡法
High-pressure microscopy for manipulating cellular architecture and function
○西山 雅祥（京大 白眉セ）
Masayoshi Nishiyama (*The HAKUBI Center, Kyoto Univ.*)

3SFA-05 集光レーザー摂動を用いた神経回路網における分子ダイナミクスの直接操作
Direct manipulation of molecular dynamics in neuronal network with laser-induced perturbation
○細川 千絵（産総研・バイオメディカル）
Chie Hosokawa (*Biomed. Res. Inst., AIST*)

3SFA-06 *Xenopus* 卵抽出液を封入した小胞中でのアクチンの流れと小胞運動
Actin flows in *Xenopus* egg extract confined in oil and generates a force for migration of the extract
○野田 直紀, 馬渕 一誠（学習院大学・理・生命）
Naoki Noda, Issei Mabuchi (*Grad. Sc. Sci., Gakushuin Univ.*)

おわりに
馬渕 一誠
Issei Mabuchi

9:00～12:00　G 会場（レクチャーホール）／Room G (Lecture Hall)

3SGA　理論と実験の統合的アプローチが解き明かす生体秩序構造　- 分子から組織まで -
Towards understanding origins of order through integrated approach of experiments and theory -From molecules to tissue-

オーガナイザー：鳥澤 嵩征（情報通信研究機構），谷口 大相（明治大学）
Organizers: Takayuki Torisawa (NICT), Daisuke Taniguchi (Meiji University)

In recent years, integrated approaches of experiment and theory have become popular for understanding the underlying mechanism of order formations in biological systems where cytoskeletal elements play a central role. In this session, based on the latest findings and key ideas in a wide variety of studies ranging from pure in vitro to multicellular systems, we will discuss the integrated approaches to understand complex biological structures organized by cytoskeletons.

Opening remarks

3SGA-01 自己集積的に形成するキネシン・微小管の収縮性ネットワーク：工学応用に向けて
Self-organized Contractile Networks of Microtubules and Engineered Kinesins: Towards Engineering Applications
○平塚 祐一[1], 新田 高洋[2]（[1]北陸先端大・マテリアル, [2]岐阜大・工・応用物理コース）
Yuichi Hiratsuka[1], Takahiro Nitta[2] ([1]*Sch. Mat. Sci., JAIST*, [2]*Appl. Phys. Course, Gifu Univ.*)

3SGA-02 Formation and rupture of a motorized cytoskeletal network
Takayuki Torisawa[1,2], Daisuke Taniguchi[2,3] ([1]*Advanced ICT Research Institute, NICT*, [2]*CREST, JST*, [3]*Dept. of Physics, School of Science and Technology, Meiji Univ.*)

3SGA-03 神経幹細胞のネマチックパターン
Active nematics of collective neural stem cells
○川口 喬吾[1], 影山 龍一郎[2], 佐野 雅己[3]（[1]ハーバード医大, [2]京大ウィルス研, [3]東大理物理）
Kyogo Kawaguchi[1], Ryoichiro Kageyama[2], Masaki Sano[3] ([1]*Dept. Syst. Biol., Harvard Med. School*, [2]*Inst. for Virus Res., Kyoto Univ.*, [3]*Dept. Phys., Univ. Tokyo*)

3SGA-04 Inference for the mechanics of moving cell sheets
Yohei Kondo[1], Kazuhiro Aoki[2], Shin Ishii[1] ([1]*Grad. Sch. Info., Kyoto Univ.*, [2]*Grad. Sch. Med., Kyoto Univ.*)

3SGA-05 線虫 C. elegans における減数分裂期細胞質流動の自己組織化の機構
A mechanism of self-organization in meiotic cytoplasmic streaming of the C. elegans embryo
木村 健二[1,2], ○木村 暁[1,2]（[1]遺伝研・細胞建築, [2]総研大・遺伝学専攻）
Kenji Kimura[1,2], **Akatsuki Kimura**[1,2] ([1]*Cell Arch. Lab., Nat. Inst. Genetics*, [2]*Dept. Genetics, SOKENDAI*)

9:00～12:00　H 会場（大講義室 A）／Room H (Lecture Room A)

3SHA　新学術領域研究「少数性生物学ー個と多数の狭間が織りなす生命現象の探求ー」共催

少数分子が担う生命現象

Biological events operated by small number of biomolecules

オーガナイザー：永井 健治（大阪大学），石島 秋彦（大阪大学）

Organizers: Takeharu Nagai (Osaka University), Akihiko Ishijima (Osaka University)

The macroscopic biological behavior has been supposed to be generated by the stochastic reactions composed of many biological molecules. However, it has been clear that a few molecules could affect the physiological phenomena, such as control of the biological rhythm by a few transcription factor and outbreak of illnesses by infection of a few virus. Here we would like to discuss how to investigate such phenomena, and also technics to be required.

概要説明
永井 健治
Takeharu Nagai

3SHA-01　インフルエンザウイルス感染と宿主細胞侵入時に惹起される細胞内シグナルの可視化
Visualisation of molecular events during influenza virus entry and infection
○大場 雄介[1], 藤岡 容一朗[1], 田端 和仁[2], 西出 真也[2], 南保 明日香[1], 野地 博行[2]（[1]北大・院医, [2]東大・院工）
Yusuke Ohba[1], Yoichiro Fujioka[1], Kazuhito V. Tabata[2], Shinya Nishide[2], Asuka Nanbo[1], Hiroyuki Noji[2] ([1]*Hokkaido Univ. Grad. Sch. Med.*, [2]*Grad. Sch. Eng., Univ. Tokyo*)

3SHA-02　*in vitro* 系で明らかになった細菌べん毛形成の分子機構とその制御
Molecular mechanism of the flagellar biogenesis revealed by *in vitro* transport assay system
○今田 勝巳（阪大・院理・高分子）
Katsumi Imada (*Grad. Sch. Sci., Osaka Univ.*)

3SHA-03　真核細胞の走化性における濃度勾配センシングと方向性のある細胞運動
Gradient sensing and directed cell migration in eukaryotic chemotaxis
○上田 昌宏[1,2]（[1]大阪大学大学院 理学研究科, [2]理化学研究所 生命システム研究センター）
Masahiro Ueda[1,2] ([1]*Graduate School of Sciences, Osaka University*, [2]*QBiC, RIKEN*)

3SHA-04　勾配感知におけるノイジーなシグナルの時間微分
Temoporal Differentiation of Noisy Signal in Gradient Sensing
○小林 徹也[1], 横田 亮[1,2]（[1]東京大学生産技術研究所, [2]複雑生命システム動態研究教育拠点）
Tetsuya Kobayashi[1], Ryo Yokota[1,2] ([1]*Institute of Industrial Science, University of Tokyo*, [2]*Research and Education Platform for Dynamic Living States*)

3SHA-05　マイノリティージェノタイプ・細胞数分布を 1 細胞レベルで同定・定量する新技術
A new method for identification of minor genotype and measurement of cell-number distribution at the single cell level
○城口 克之（理研・統合生命医）
Katsuyuki Shiroguchi (*IMS RIKEN*)

3SHA-06　個体レベルの「時間」の理解に向けて
―全身・全脳透明化の先に見えるもの―
Towards System-level Understanding of Biological Time
○上田 泰己[1,2]（[1]東京大学, [2]理化学研究所　）
Hiroki R. Ueda[1,2] ([1]*The University of Tokyo*, [2]*RIKEN, QBiC*)

総合討論
石島 秋彦, 永井 健治
Akihiko Ishijima, **Takeharu Nagai**

9:00～12:00　I会場（大講義室B）／Room I (Lecture Room B)

3SIA　タンパク質を活かす多成分システムの分子レベル解析：水和効果から細胞内環境へ
Protein structures and functions in multi-component systems: From hydration to intracellular environment

オーガナイザー：吉村 成弘（京都大学），松林 伸幸（大阪大学）
Organizers: Shigehiro Yoshimura (Kyoto University), Nobuyuki Matubayasi (Osaka University)

It is well recognized that protein structures and functions are strongly affected by the surrounding environments such as water and lipid membrane. The next challenge is then to quantitatively analyze the interactions of protein with the surroundings toward possible tuning of hydration effects and molecular-level understanding of intracellular environment. These can be achieved through molecular analysis of proteins in multi-component systems. In the present symposium, we introduce and discuss recent progresses of theoretical and experimental studies related to proteins in multi-component system.

はじめに

3SIA-01　タンパク質の分子間相互作用への溶媒効果
Solvation effects on protein interaction with other molecules
○北尾 彰朗（東大・分生研）
Akio Kitao (*IMCB, Univ. Tokyo*)

3SIA-02　Time-resolved resonance Raman observation of proteins in action
Yasuhisa Mizutani (*Osaka Univerisity*)

3SIA-03　タンパク質構造に対する環境効果のエネルギー相関解析
Correlation Analysis of Environmental Effect on Protein Structure with Explicit Solvent
○松林 伸幸（大阪大学 大学院基礎工学研究科 化学工学領域）
Nobuyuki Matubayasi (*Division of Chemical Engineering, Graduate School of Engineering Science, Osaka University*)

3SIA-04　極限環境生物がデザインした蛋白質
Proteins designed by extremophiles
○三本木 至宏（広島大学）
Yoshihiro Sambongi (*Hiroshima University*)

3SIA-05　ヘリカルリピートタンパク質の細胞内での構造と機能
Intracellular structure and function of helical repeat proteins
○吉村 成弘, 小西 秀明, 浅井 賢（京大・院生命科学）
Shigehiro Yoshimura, Hide Konishi, Suguru Asai (*Grad. Sch. Biostudies, Kyoto Univ.*)

3SIA-06　バクテリア細胞質中の蛋白質および代謝物のダイナミクス・安定性・相互作用：全原子分子動力学法による理論的研究
Dynamics, Stability, and Interactions of Proteins and Metabolites in Bacterial Cytoplasm: All-atom Molecular Dynamics Study
○優 乙石[1,4], 森 貴治[1], 安藤 格士[2], 原田 隆平[3], Jung Jaewoon[3], 杉田 有治[1,2,3,4], Feig Michael[5]（[1]理研 杉田理論分子科学研究室, [2]理研 生命システム研究センター, [3]理研 計算科学研究機構, [4]理研 理論科学連携研究推進グループ, [5]ミシガン州立大学 生化学・分子生物学科）
Isseki Yu[1,4], Takaharu Mori[1], Tadashi Ando[2], Ryuhei Harada[3], Jaewoon Jung[3], Yuji Sugita[1,2,3,4], Michael Feig[5] ([1]*RIKEN Theoretical Molecular Science Laboratory*, [2]*RIKEN QBiC*, [3]*RIKEN AICS*, [4]*RIKEN iTHES*, [5]*Department of Biochemistry and Molecular Biology, Michigan State University*)

3SIA-07　NMR を使ったジスルフィド結合の細胞内解析
Stability of disulfide bonds of proteins in the cytosolic space analyzed using NMR spectroscopy
村山 秀平[2], 榎園 能章[1], 赤木 謙一[3], 関山 直孝[1], 猪股 晃介[4], 白川 昌宏[2], ○杤尾 豪人[1]（[1]京都大学大学院理学研究科, [2]京都大学大学院工学研究科, [3]医薬基盤研, [4]理化学研究所）
Shuhei Murayama[2], Yoshiaki Enokizono[1], Ken-ichi Akagi[3], Naotaka Sekiyama[1], Kohsuke Inomata[4], Masahiro Shirakawa[2], **Hidehito Tochio**[1] ([1]*Graduate School of Science, Kyoto University*, [2]*Graduate School of Engineering, Kyoto University*, [3]*National Institute of Biomedical Innovation*, [4]*RIKEN*)

おわりに

9:00〜12:00　J 会場（105 教室）／Room J (Room 105)

3SJA　生体分子におけるレアイベントの探求
Exploring rare events in biomolecular systems

オーガナイザー：藤崎 弘士（日本医科大学），米澤 康滋（近畿大学）
Organizers: Hiroshi Fujisaki (Nippon Medical School), Yasushige Yonezawa (Kinki University)

Biomolecules consist of several different types of atoms, forming anisotropic bonded and non-bonded interactions. This heterogeneous nature leads to significant complexity in their configurations and dynamics. When a biomolecule plays a functional role, some molecular motions including conformational changes are often induced by some stimuli, and the biomolecule explores many substates (metastable states) during conformational change, which can be rare in terms of molecular timescales. In this symposium, both computational chemists and experimentalists investigate the rare events and substates in biomolecules and present their most recent results for discussions.

はじめに
藤崎 弘士, 米澤 康滋
Hiroshi Fujisaki, **Yasushige Yonezawa**

3SJA-01　ストリング法を用いた多剤排出トランスポーター AcrB の薬剤排出機構の解析
Drug extrusion mechanism of multidrug exporter AcrB studied by the string method
○松永 康佑（理研・AICS）
Yasuhiro Matsunaga (*RIKEN AICS*)

3SJA-02　大規模分子集合体系におけるレアイベントの分子動力学計算
Molecular dynamics study of rare events of large-scale molecular systems
○吉井 範行（名大院工）
Noriyuki Yoshii (*Grad. Sch. Eng., Nagoya Univ.*)

3SJA-03　緩和モード解析による蛋白質の動的性質の研究
Exploring Dynamics and Kinetics of Proteins using Relaxation Mode Analysis
○光武 亜代理[1,2]（[1]慶應大・理工・物理, [2]さきがけ）
Ayori Mitsutake[1,2] ([1]*Dep. of Phys., Keio Univ.*, [2]*JST, Presto*)

3SJA-04　生物学的レアイベントを再現する効率的構造サンプリング手法
Simple yet powerful conformational sampling methods for reproducing biologically rare events
○原田 隆平（筑波大学計算科学研究センター）
Ryuhei Harada (*CCS, Univ. of Tsukuba*)

3SJA-05　時間分解共鳴ラマン分光法をもちいたレチナールタンパク質におけるレアイベント観測
Observation of rare events in retinal proteins revealed by time-resolved resonance Raman spectroscopy
○水野 操（大阪大学大学院理学研究科）
Misao Mizuno (*Graduate School of Science, Osaka University*)

3SJA-06　天然変性領域にレアに生じる構造が持つ役割
Role of rarely happening fold in intrinsically disordered proteins (IDPs)
○楯 真一[1,2]（[1]広島大・院理・数理分子, [2]広島大・クロマチン動態数理）
Shin-ichi Tate[1,2] ([1]*Dept. Math. and Life Sci., Hiroshima Univ.*, [2]*RcMcD, Hiroshima Univ.*)

9:00～12:00　K 会場（107 教室）／Room K (Room 107)

3SKA　人工細胞を創る・動かす・活用する
Artificial cells: Preparation, application, and activation

オーガナイザー：藤原 慶（慶應義塾大学），森田 雅宗（東京工業大学）
Organizers: Kei Fujiwara (Keio University), Masamune Morita (Tokyo Institute of Technology)

Recent progress has developed methods to prepare life-mimicking liposomes (artificial cells) as platforms to analyze chemical and physical aspects of living cells in vitro. Recently, dynamic functions such as taxis and evolvability are installed in these artificial cells. Can artificial cells be or go beyond living cells? In this symposium, relatively young speakers in these research fields talk their recent achievements and will discuss the future of artificial cells.

オープニング
森田 雅宗
Masamune Morita

3SKA-01　人工細胞構築のための細胞サイズリポソームの生成と応用
Preparation and application of cell-sized liposomes for synthesis of artificial cells
○森田 雅宗[1,2]（[1]東工大・院総理工, [2]学振特別研究員）
Masamune Morita[1,2] ([1]*Interdisciplinary Grad. Sch. Sci. and Eng., Tokyo Tech.*, [2]*JSPS Research Fellow*)

3SKA-02　ボトムアップアプローチによる人工細胞の作製
Preparation of artificial cell models by bottom-up approach
○神谷 厚輝[1,2]（[1]神奈川科学技術アカデミー, [2]JST さきがけ）
Koki Kamiya[1,2] ([1]*Kanagawa Academy of Science and Technology*, [2]*PRESTO, JST*)

3SKA-03　細胞サイズ閉鎖空間内でのアクチン細胞骨格の再構成
Reconstitution of actin cytoskeleton in a cell-sized confined space
○宮崎 牧人[1], 千葉 雅隆[1], 江口 宙輝[1], 大木 高志[1], 石渡 信一[1,2]（[1]早稲田大・物理, [2]早稲田バイオサイエンスシンガポール研究所）
Makito Miyazaki[1], Masataka Chiba[1], Hiroki Eguchi[1], Takashi Ohki[1], Shin'ichi Ishiwata[1,2] ([1]*Dept. of Physics, Waseda Univ.*, [2]*WABIOS, Waseda Univ.*)

3SKA-04　ミクロ水滴から出発する自己駆動型人工細胞へのアプローチ
Approach to self-propelled artificial cells from microdroplets
○伊藤 弘明, 市川 正敏（京大・院理）
Hiroaki Ito, Masatoshi Ichikawa (*Grad. Sch. Sci., Kyoto Univ.*)

3SKA-05　人工細胞を活用した高分子ミクロゲル形状の物理的な制御
Shape control of biopolymer microgels utilizing model cells
○柳澤 実穂（東京農工大学大学院工学研究院先端物理工学部門）
Miho Yanagisawa (*Dept. Appl. Phys.,Tokyo Univ. of Agri. & Tech.*)

3SKA-06　膜タンパク質を人工細胞に組み込み、進化させる
Installation and directed evolution of membrane proteins in artificial cells
○藤井 聡志（大阪大・院情報科学）
Satoshi Fujii (*Grad. Info. Sci., Osaka Univ.*)

3SKA-07　細胞再構成へ：生細胞に近い人工細胞の創成
Creating Life-mimicking Artificial Cells toward re-building living cells
○藤原 慶（慶應義塾大学理工学部生命情報学科）
Kei Fujiwara (*Dept. Biosciences and Informatics, Keio University*)

3SKA-08　合成細胞生物学ツールとして利用可能な GUV 人工細胞モデル
GUV-based artificial cell model available as constructive cell biology tool
○野村 慎一郎（東北大学 工学研究科 バイオロボティクス専攻）
Shin-ichiro Nomura (*Department of Bioeng. Robotics., Tohoku Univ.*)

クロージング
藤原 慶
Kei Fujiwara

1C1320　アミロイド β1-42 多量体の構造ダイナミクスの高速 AFM 観察
Video imaging of structural dynamics of individual amyloid β 1-42 aggregates
○伊丹 将大[1], 小野 賢二郎[2], 中山 隆宏[3]（[1]金沢大・理工, [2]金沢大学病院, [3]金沢大・AFM）
Masahiro Itami[1], Kenjiro Ono[2], Takahiro Nakayama[3] ([1]*Grad. Sch. Sci., Univ. Kanazawa*, [2]*Kanazawa Univ. Hospital*, [3]*Kanazawa Univ. Bio AFM FRC*)

1C1335　ユビキチンリガーゼ (HECT 型 E3) のユビキチン化に伴う動態の高速 AFM 観察
Observation of the dynamics associated with ubiquitination of HECT E3 ubiquitin ligase using High speed AFM
○小林 史典[1], 春山 隆充[2], 中山 隆宏[2], 古寺 哲幸[2], 紺野 宏記[2]（[1]金沢大・理工, [2]金沢大・AFM）
Fuminori Kobayashi[1], Takamitsu Haruyama[2], Takahiro Nakayama[2], Noriyuki Kodera[2], Hiroki Konno[2] ([1]*Grad. Sch. Sci., Univ. Kanazawa*, [2]*Kanazawa Univ. Bio AFM FRC*)

1C1350　高速 AFM による抗体のやわらかさ測定
High-Speed AFM reveals swinging nature of antibody with flexible arms
○小谷 則遠, ラマヌジャム クマレサン, 川元 洋子, 岡田 孝夫（株式会社 生体分子計測研究所）
Norito Kotani, Kumaresan Ramanujam, Yoko Kwamoto, Takao Okada (*Research Institute of Biomolecule Metrology Co.,Ltd.*)

1C1405　高速原子間力顕微鏡の温度制御機構の開発と好熱菌 FliI の観察
Development of Temperature Controlled High-Speed AFM and Observation of Thermus Thermophilus FliI
○足立 彗[1], 内橋 貴之[1], 今田 勝己[2], 横山 謙[3], 安藤 敏夫[1]（[1]金沢大理工, [2]阪大理, [3]京産大総合生命）
Kei Adachi[1], Takayuki Uchihashi[1], Katsumi Imada[2], Ken Yokoyama[3], Toshio Ando[1] ([1]*Coll. Sci. & Eng., Kanazawa Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Facul. Biosci., Kyoto Sangyo Univ.*)

休憩 14:20-14:25

1C1425　ヘテロな系での AFM の応用に向けた AFM・TEM の相関顕微鏡法
Correlative Atomic Force and Electron Microscopy toward Applications of Atomic Force Microscopy to Heterogeneous Systems
○山田 裕太郎[1], 春山 隆充[1], 紺野 宏記[1], 島袋 勝弥[1]（[1]宇部工業高等専門学校, [2]AFMセンター）
Yutaro Yamada[1], Takamitsu Haruyama[1], Hiroki Konno[1], Katsuya Shimabukuro[1] ([1]*UNCT*, [2]*Bio-AFM, Kanazawa Univ.*)

1C1440　大球コロイドの周りに分布している小球コロイドの数密度分布の計測理論
Measurement theory of density distribution of small colloids around a large colloid
○天野 健一, 橋本 康汰, 西 直哉, 作花 哲夫（京都大学大学院工学研究科）
Ken-ichi Amano, Kota Hashimoto, Naoya Nishi, Tetsuo Sakka (*Graduate School of Engineering, Kyoto University*)

1C1455　ナノスケールの形状・化学物質濃度プロファイルを可視化するナノ電気化学顕微鏡の創成
Development of Nano Electrochemical Microscopy for Visualizing Nanoscale Cell Surface Topography and Chemical Profile
○高橋 康史[1,2,3], 井田 大貴[2], 珠玖 仁[2], 末永 智一[1,2]（[1]東北大WPI－AIMR, [2]東北大院環境, [3]JST さきがけ）
Yasufumi Takahashi[1,2,3], Hiroki Ida[2], Hitoshi Shiku[2], Tomokazu Matsue[1,2] ([1]*WPI-AIMR of Tohoku University*, [2]*Environmental studies, Graduate school of Tohoku University*, [3]*JST PREST*)

1C1510　Spectral fingerprinting of individual cells observed by cavity-reflection-enhanced light-absorption microscopy
Yoshiyuki Arai[1], Takayuki Yamamoto[1], Takeo Minamikawa[2], Tetsuro Takamatsu[2], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*Grd. Sch. Med Sci., Kyoto Pref. Univ.*)

休憩 15:25-15:30

1C1530　細胞観察に向けたティップスキャン型高速 AFM の改良
Improvement of tip-scan HS-AFM for live-cell imaging
○山中 信之介[1], 渡辺 大輝[1,2], 内橋 貴之[1,3], 安藤 敏夫[1,3]（[1]金大院・自然科学研究科, [2]生体分子計測研究所, [3]バイオAFMセンター）
Shin-nosuke Yamanaka[1], Hiroki Watanabe[1,2], Takayuki Uchihashi[1,3], Toshio Ando[1,3] ([1]*Grad. Sch. Sci, Univ. Kanazawa*, [2]*RIBM*, [3]*Bio-AFM Center*)

1C1545　Long-tip 高速原子間力顕微鏡による生きた細胞の形態観察
Live-cell imaging by long-tip high-speed atomic force microscopy
○柴田 幹大[1,2], 内橋 貴之[2,3], 安藤 敏夫[2,3], 安田 涼平[1]（[1]マックスプランクフロリダ, [2]金沢大・理工, [3]金沢大・バイオAFM）
Mikihiro Shibata[1,2], Takayuki Uchihashi[2,3], Toshio Ando[2,3], Ryohei Yasuda[1] ([1]*MPFI*, [2]*Dept. Phys., Kanazawa Univ.*, [3]*Bio-AFM, Kanazawa Univ.*)

1C1600　動物細胞の 1 細胞系譜の取得に向けたマイクロ流体デバイス

Microfluidic device for tracking mammalian cells along single-cell lineages

○清田 晃央[1], 若本 祐一[1,2]（[1]東京大学総合文化研究科広域科学専攻, [2]複雑系生命システム研究センター）

Akihisa Seita[1], Yuichi Wakamoto[1,2] ([1]*Department of Basic Science Graduate, School of Arts and Science, University of Tokyo*, [2]*Research Center for Complex Systems Biology*)

13:20〜16:15　D 会場（103 教室）／Room D (103)
1D　光生物：視覚・光受容 I / Photobiology: Vision & Photoreception I

1D1320　単量体・二量体平衡の定量的解析による Photozipper の分子機構解明

Quantitative analyses of the monomer-dimer equilibrium reveal the molecular mechanism of Photozipper

○中谷 陽一, 久冨 修（大阪大・院理学）

Yoichi Nakatani, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

1D1335　(6–4)光回復酵素の拡張された電子移動経路

An expanded electron transfer pathway in the (6-4) photolyase

○山元 淳平[1], Müller Pavel[2], 清水 幸平[1], Brettel Klaus[2], 岩井 成憲[1]（[1]大阪大学大学院基礎工学研究科, [2]CEA Saclay, France）

Junpei Yamamoto[1], Pavel Müller[2], Kohei Shimizu[1], Klaus Brettel[2], Shigenori Iwai[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*CEA Saclay, France*)

1D1350　CPD 光回復酵素と(6-4)光回復酵素の機能転換

Functional conversion of CPD and (6-4) photolyases by mutation

○山田 大智[1], Dokainish Hisham M.[2], 岩田 達也[1], 山元 淳平[3], 石川 智子[4], 藤堂 剛[4], 岩井 成憲[3], Getzoff Elizabeth D.[5], 北尾 彰朗[2], 神取 秀樹[1]（[1]名工大・院工, [2]東大・分子細胞生物学研, [3]阪大院・基礎工, [4]阪大・院医, [5]米国・スクリプス研）

Daichi Yamada[1], Hisham M. Dokainish[2], Tatsuya Iwata[1], Junpei Yamamoto[3], Tomoko Ishikawa[4], Takeshi Todo[4], Shigenori Iwai[3], Elizabeth D. Getzoff[5], Akio Kitao[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*IMCB, Univ. Tokyo*, [3]*Grad. Sch. Eng. Sci., Osaka Univ.*, [4]*Grad. Sch. Med., Osaka Univ.*, [5]*The Scripps Res. Inst., USA*)

1D1405　シロイヌナズナクリプトクロム 1 の光反応における赤外分光研究

FTIR study of the Arabidopsis Cryptochrome1 photoreaction

○三國 克紘[1], 山田 大智[1], 岩田 達也[1], 人見 研一[2], Getzoff Elizabeth D.[2], 神取 秀樹[1]（[1]名工大・院工, [2]米国スクリプス研）

Katsuhiro Mikuni[1], Daichi Yamada[1], Tatsuya Iwata[1], Kenichi Hitomi[2], Elizabeth D. Getzoff[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*The Scripps Res. Inst. USA*)

休憩 14:20-14:25

1D1425　視物質の低い熱活性化頻度をもたらす分子メカニズム

Molecular mechanism of the low thermal activation rate of visual pigments

○小島 慧一[1], 柳川 正隆[2], 山下 高廣[1], 松谷 優樹[1], 今元 泰[1], 松山 オジョス武[3], 中西 香爾[4], 山野 由美子[5], 和田 昭盛[5], 佐甲 靖志[2], 七田 芳則[1]（[1]京大・院理, [2]理研・細胞情報, [3]理研・CDB, [4]コロンビア大, [5]神戸薬科大学）

Keiichi Kojima[1], Masataka Yanagawa[2], Takahiro Yamashita[1], Yuki Matsutani[1], Yasushi Imamoto[1], Take Matsuyama[3], Koji Nakanishi[4], Yumiko Yamano[5], Akimori Wada[5], Yasushi Sako[2], Yoshinori Shichida[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Cell. Info. Lab., RIKEN*, [3]*CDB, RIKEN*, [4]*Columbia Univ.*, [5]*Kobe Pharm. Univ.*)

1D1440　レチナール異性化によるロドプシン活性化のメカニズムの解析

Mechanism of how retinal isomerization changes the structure of rhodopsin to the active state

○木股 直規[1], Sheves Mordechai[2], Reeves Philip[3], Smith Steven[1]（[1]Stony Brook大・生化学, [2]Weizmann研・有機化学, [3]Essex大・生物学）

Naoki Kimata[1], Mordechai Sheves[2], Philip Reeves[3], Steven Smith[1] ([1]*Dept. Biochem., Stony Brook Univ.*, [2]*Dept. Organic Chem., Weizmann Inst.*, [3]*Dept. Biol. Sci., Univ. Essex*)

1D1455　青感受性視物質における Y265 の役割

Role of Y265 in blue-sensitive visual pigment

○野中 祐貴[1], 片山 耕大[1,2], 筒井 圭[3], 今井 啓雄[3], 神取 秀樹[1]（[1]名工大院工, [2]ケースウェスタンリザーブ大, [3]京大霊長研）

Yuki Nonaka[1], Kota Katayama[1,2], Kei Tsutsui[3], Hiroo Imai[3], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Dept. Pharm., CWRU, USA*, [3]*Primate Res. Inst., Kyoto Univ.*)

1D1510　サル緑感受性視物質に対する陰イオン効果の構造研究

Anion effect to monkey green studied by light-induced difference FTIR spectroscopy

○中村 駿太[1], 片山 耕大[1,2], 今井 啓雄[3], 神取 秀樹[1]（[1]名工大, [2]ケース・ウェスタン・ブリーズ大, [3]京大 霊長研）

Shunta Nakamura[1], Kota Katayama[1,2], Hiroo Imai[3], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Dept. Pharm., CWRU, USA*, [3]*Primate Res. Inst., Kyoto Univ.*)

休憩 15:25-15:30

1D1530 **オプシン発現培養細胞の生化学的な応答に基づく非視覚型オプシンの分光感度の推定**

Estimating spectral sensitivities of non-visual opsins based on biochemical responses of opsin-expressing cultured cells

○杉原 智博[1], 永田 崇[1], 小柳 光正[1,2,3], 寺北 明久[1,2]（[1]大阪市立大・院理, [2]複合先端, [3]JST・さきがけ）

Tomohiro Sugihara[1], Takashi Nagata[1], Mitsumasa Koyanagi[1,2,3], Akihisa Terakita[1,2] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*OCARINA*, [3]*JST-PRESTO*)

1D1545 **TMT1 オプシンと TMT2 オプシンの分子特性比較解析**

Comparative studies on the molecular properties between TMT1 and TMT2 opsins

○酒井 佳寿美, 山下 高廣, 今元 泰, 七田 芳則（京都大・院理）

Kazumi Sakai, Takahiro Yamashita, Yasushi Imamoto, Yoshinori Shichida (*Grad. Sch. Sci., Kyoto Univ.*)

1D1600 **G_s タンパク質の光制御に向けたキメラタンパク質の創出**

Construction of chimeric proteins for optical control of G_s-protein activity

○吉田 一帆[1], 井上 圭一[1,2], 山下 高廣[3], 吉住 玲[1], 田中 瑞奈[1], 佐々木 賢吾[1], 七田 芳則[3], 神取 秀樹[1]（[1]名工大・院工, [2]JSTさきがけ, [3]京大・院理）

Kazuho Yoshida[1], Keiichi Inoue[1,2], Takahiro Yamashita[3], Rei Abe-Yoshizumi[1], Mizuna Tanaka[1], Kengo Sasaki[1], Yoshinori Shichida[3], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*, [3]*Grad. Sch. Sci., Univ. Kyoto*)

13:20〜16:15　E 会場（104 教室）／Room E (104)
1E　蛋白質：構造・構造機能相関 II / Proteins: Structure, Structure-function relationship II

1E1320 **GTP 加水分解と PKC リン酸化によるダイナミン–コルタクチン複合体の制御**

Regulation of Dynamin-Cortactin complex by GTP hydrolysis and PKC phosphorylation

○竹居 孝二[1,4], 阿部 匡史[1,4], 熊谷 祐介[2], 宮垣 祐志[1], 竹田 哲也[1,4], 内橋 貴之[2,3,4], 安藤 敏夫[2,3,4], 山田 浩司[1]（[1]岡山大・院医歯薬, [2]金沢大・理工学域, [3]金沢大・バイオAFM先端研究センター, [4]科学技術振興機構戦略的創造研究推進事業）

Kohji Takei[1,4], Tadashi Abe[1,4], Yusuke Kumagai[2], Yuji Miyagaki[1], Tetsuya Takeda[1,4], Takayuki Uchihashi[2,3,4], Toshio Ando[2,3,4], Hiroshi Yamada[1] ([1]*Okayama University*, [2]*Kanazawa Univ. College Sci. & Engineering*, [3]*Kanazawa Univ. Bio-AFM Frontier Res. Ctr*, [4]*CREST, Japan Science and Technology Agency (JST)*)

1E1335 **高速原子間力顕微鏡によるペルオキシレドキシン高分子量複合体の観察**

Direct visualization of high molecular weight complex of peroxiredoxin using high-speed AFM

○春山 隆充[1], 内橋 貴之[1,2], 紺野 宏記[1]（[1]金沢大・理工・バイオAFMセンター, [2]金沢大・理工）

Takamitsu Haruyama[1], Takayuki Uchihashi[1,2], Hiroki Konno[1] ([1]*Bio-AFM FRC, Coll. Sci. & Eng., Kanazawa Univ.*, [2]*Coll. Sci. & Eng., Kanazawa Univ.*)

1E1350 **高速原子間力顕微鏡による Kai タンパク質間の相互作用の観察**

High-speed AFM observation of dynamic interactions between Kai proteins

○杉山 翔吾[1], 盛 哲也[2], Carl H. Johnson[2], 内橋 貴之[1,3]（[1]金沢大院自然科学, [2]Dept.of Biol.Sci.,Vanderbilt.Univ., [3]バイオAFM先端研究センター）

Shogo Sugiyama[1], Tetsuya Mori[2], Johnson Carl H.[2], Takayuki Uchihashi[1,3] ([1]*Dept. of Phys., Univ. Kanazawa*, [2]*Dept. of Biol. Sci., Vanderbilt. Univ.*, [3]*Bio-AFM FRC., Univ. Kanazawa*)

1E1405 **Development of structural analysis system of protein-protein complexes based on identification of hydrogen bond network**

Masaru Tateno, Takuya Takeda, Jiyoung Kang (*Grad. Sch. Life Sci., Univ. Hyogo*)

休憩 14:20-14:25

1E1425 **効率よくウイルス DNA に変異を導入する APOBEC3G の脱アミノ化機構**

The deamination mechanism of APOBEC3G required for effective gene mutation in viral genome

○神庭 圭佑[1,2], 永田 崇[1,2], 片平 正人[1,2]（[1]京大・エネルギー理工学研究所, [2]京大・エネルギー科学）

Keisuke Kamba[1,2], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Inst. of Advanced Energy, Kyoto Univ.*, [2]*Grad. Sch. of Energy Science, Kyoto Univ.*)

1E1440 **Structural modeling of negatively supercoiled DNA recognition peptide complexed with crossover DNA**

Kakeru Sakabe, Jiyoung Kang, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

1E1455 **蛋白質の機能構造として働く励起構造**

An excited-state conformer acts as the functional conformer of the protein

稲葉 理美[1], 前野 覚大[2], 櫻井 一正[2], 池上 貴久[3], 赤坂 一之[1], ○織田 昌幸[1]（[1]京府大・院生命環境科学, [2]近大・高圧研, [3]横市大・院生命医科学）

Satomi Inaba[1], Akihiro Maeno[2], Kazumasa Sakurai[2], Takahisa Ikegami[3], Kazuyuki Akasaka[1], **Masayuki Oda**[1] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ.*, [2]*High Pressure Protein Res. Center, Kinki Univ.*, [3]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*)

1E1510 **過渡共鳴ラマン分光法を用いたプロテオロドプシン光反応初期中間体の解析**

Photointermediates of proteorhodopsin studied by transient resonance Raman spectroscopy

○山領 春輝[1], 田実 真一[1], 田母神 淳[2], 加茂 直樹[2], 海野 雅司[1]（[1]佐賀大学, [2]松山大学）

Haruki Yamaryo[1], Shinichi Tajitsu[1], Jun Tamogami[2], Naoki Kamo[2], Masashi Unno[1] ([1]*Saga University*, [2]*Matsuyama University*)

休憩 15:25-15:30

1E1530 **抗ニトロフェニル抗体の鍵となるアミノ酸残基 V_H33 の抗原結合における役割**

Role of the key residue at V_H33 of anti-nitrophenyl antibody in its antigen binding

○佐藤 優穂[1], 丸野 孝浩[2], 深田 はるみ[3], 小林 祐次[2], 東 隆親[4], 織田 昌幸[1]（[1]京府大・院生命環境科学, [2]阪大・院工, [3]阪府大・院生命環境科学, [4]東理大・生命研）

Yusui Sato[1], Takahiro Maruno[2], Harumi Fukada[3], Yuji Kobayashi[2], Takachika Azuma[4], Masayuki Oda[1] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ.*, [2]*Grad. Sch. of Eng., Osaka Univ.*, [3]*Grad. Sch. of Life and Environ. Sci., Osaka Pref. Univ.*, [4]*Res. Ins. for Biol. Sci., Tokyo Univ. of Sci.*)

1E1545 **抗体の新たな抗原認識機構がもたらす特異性創出原理**

New paradigm for antibody-antigen recognition enabling extraordinarily high specificity

○服部 峰充[1], Lai Darson[1], Dementieva Irina[1], Montano Sherwin[1], Kurosawa Kohei[1], Koide Akiko[1], Ruthenburg Alexander J.[1,2], 小出 昌平[1]（[1]Dept. of Biochem. and Mol. Biol., The Univ. of Chicago, [2]Dept. of Mol. Genetics and Cell Biol., The Univ. of Chicago）

Takamitsu Hattori[1], Darson Lai[1], Irina Dementieva[1], Sherwin Montano[1], Kohei Kurosawa[1], Akiko Koide[1], Alexander J. Ruthenburg[1,2], Shohei Koide[1] ([1]*Dept. of Biochem. and Mol. Biol., The Univ. of Chicago*, [2]*Dept. of Mol. Genetics and Cell Biol., The Univ. of Chicago*)

1E1600 **Regulation of Adaptive Immunity: Activation and Inhibition of the ZAP-70 Kinase Domain**

Roland G. Huber[1], Hao Fan[1,2], Peter J. Bond[1,2] ([1]*Bioinformatics Institute, A*STAR*, [2]*Department of Biological Sciences, NUS*)

13:20〜16:15　J 会場（105 教室）／Room J (105)
1J　細胞生物的課題 I / Cell biology I

1J1320 **ダイナミンによる膜切断メカニズムの高速 AFM イメージング解析**

Pinch or Pop: HS-AFM imaging analyses of membrane scission mechanisms by Dynamin

○竹田 哲也[1,4], 熊谷 祐介[2], 背山 佳穂[1], 楊 恵然[1], 山田 浩司[1,4], 田岡 東[2,3], 内橋 貴之[2,3,4], 竹居 孝二[1,4], 安藤 敏夫[2,3,4]（[1]岡山大・医歯薬, [2]金沢大・理工, [3]金沢大・バイオAFM, [4]JST・CREST）

Tetsuya Takeda[1,4], Yusuke Kumagai[2], Kaho Seyama[1], Huiran Yang[1], Hiroshi Yamada[1,4], Azuma Taoka[2,3], Takayuki Uchihashi[2,3,4], Kohji Takei[1,4], Toshio Ando[2,3,4] ([1]*Grad. Sch. Med. Dent. Pharma. Sci., Okayama Univ.*, [2]*Coll. Sci. Eng., Kanazawa Univ.*, [3]*Bio AFM, Kanazawa Univ.*, [4]*CREST, JST*)

1J1335 **Multi-state transitions of PTEN mediate spontaneous signal generation and environmental bias in cell migration**

Satomi Matsuoka[1,2], Masahiro Ueda[1,2] ([1]*QBiC, Riken*, [2]*Grad. Sch. Sci., Osaka Univ.*)

1J1350 **Instantaneous fluorescence polarization microscopy for mapping position and orientation of protein assemblies in living cells with single molecule sensitivity**

Tomomi Tani[1], Shalin Mehta[1], Molly McQuilken[2], Patricia Occhipinti[2], Amitabh Verma[1], Rudolf Oldenbourg[1], Amy Gladfelter[2] ([1]*Marine Biological Laboratory, USA*, [2]*Dartmouth College, USA*)

1J1405 **大気圧電子顕微鏡 ASEM による水中深さ方向の観察：神経の細胞輸送研究や組織の癌術中迅速診断への可能性**

Depth observation of Tissues and cells in Liquid by ASEM: Applicability to Intra-Operative Cancer Diagnosis and cell trafficking study

○佐藤 主税, 海老原 達彦, Memtily Nassirhadjy, 佐藤 真理, 岡田 知子, 川田 正晃（産総研）

Chikara Sato, Tatsuhiko Ebihara, Nassirhadjy Memtily, Mari Sato, Tomoko Okada, Masaaki Kawata (*AIST*)

休憩 14:20-14:25

1J1425 **癌進行に伴うヒト胃癌細胞の形状揺らぎと接着能の変化**

Change in Shape Fluctuation and Adhesion of Human Gastric Cells Induced by Cancer Progression

○山本 暁久[1], 鶴山 竜昭[2], 田中 求[1,3]（[1]京大iCeMS, [2]京大医学部, [3]ハイデルベルグ大化学物理）

Akihisa Yamamoto[1], Tatsuaki Tsuruyama[2], Motomu Tanaka[1,3] ([1]*iCeMS, Kyoto Univ.*, [2]*Diagn. Pathol., Kyoto Univ.*, [3]*Phys. Chem., Univ. of Heidelberg*)

1J1440 **１細胞分泌実時間イメージングが明らかにした細胞分泌動態の不均一性**

Real-time single-cell secretion imaging revealed heterogeneity of the cell secretion

○白崎 善隆[1,2], 劉 霆[3], 山口 良文[3], 山岸 舞[1,2], 鈴木 信勇[1], 三浦 正幸[3], 小原 收[2], 上村 想太郎[1]（[1]東大・院理, [2]理研・IMS, [3]東大・院薬）

Yoshitaka Shirasaki[1,2], Ting Liu[3], Yoshifumi Yamaguchi[3], Mai Yamagishi[1,2], Nobutake Suzuki[1], Masayuki Miura[3], Osamu Ohara[2], Sotaro Uemura[1] ([1]*Grad. Sch. Sci., Tokyo Univ.*, [2]*IMS, RIKEN*, [3]*Grad. Sch. Pharm., Tokyo Univ.*)

1J1455 **Visualizing mechanical force transmission at integrin molecules and the interior architecture of focal adhesions with traction maps**

Masatoshi Morimatsu[1], Armen H. Mekhdjian[1], Alice C. Chang[1], Steven J. Tan[1], Alexander R. Dunn[1,2] ([1]*Department of Chemical Engineering, Stanford University*, [2]*Stanford Cardiovascular Institute, Stanford University School of Medicine*)

1J1510 **細胞膜に繋留された小胞を介した新しいシグナル変換機構：1 分子イメジングによる解明**

New signal transduction mechanism mediated by plasma-membrane-tethered vesicles: unraveling by single-molecule imaging

○廣澤 幸一朗[1], 吉田 謙太[2], 野崎 梢平[3], 角山 貴昭[2], 鈴木 健一[1,4], 中山 和久[3], 藤原 敬宏[1], 楠見 明弘[1,2]（[1]京都大・物質-細胞統合システム拠点, [2]京都大・再生研, [3]京都大・薬学研究科, [4]インド国立生命科学研究センターインド幹細胞・再生医学研究所）

Koichiro M. Hirosawa[1], Kenta J. Yoshida[2], Shohei Nozaki[3], Taka A. Tsunoyama[2], Kenichi G.N. Suzuki[1,4], Kazuhisa Nakayama[3], Takahiro K. Fujiwara[1], Akihiro Kusumi[1,2] ([1]*Inst. Integrated Cell-Material Sciences (WPI-iCeMS), Kyoto Univ.*, [2]*Inst. Frontier Medical Sciences, Kyoto Univ.*, [3]*Grad. Sch. Pharmaceutical Sciences, Kyoto Univ.*, [4]*NCBS/inStem, India*)

休憩 15:25-15:30

1J1530 **GPCR の特徴であるリガンド無しでの構成的シグナルは過渡的な GPCR ダイマーが誘起している**

Constitutive signaling without ligation characteristic with GPCRs is triggered by transient GPCR dimers

○笠井 倫志[1,2], 楠見 明弘[1,2]（[1]京大・再生研, [2]京大・WPI-iCeMS）

Rinshi Kasai[1,2], Akihiro Kusumi[1,2] ([1]*Inst. Front. Med. Sci., Kyoto Univ.*, [2]*WPI-iCeMS, Kyoto Univ.*)

1J1545 **FlhA と FliH/FliI との相互作用がべん毛フックの構築順序を巧みに制御する**

Role of the interaction between FlhA and the FliH/FliI complex in coordinating flagellar hook assembly

井上 由美[1], 木下 実紀[1], 難波 啓一[1,2], ○南野 徹[1]（[1]阪大院・生命機能, [2]理研・QBiC）

Yumi Inoue[1], Miki Kinoshita[1], Keiichi Namba[1,2], **Tohru Minamino**[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*QBiC, RIKEN*)

1J1600 **Biochemical and functional characterization of the effects of a single point mutation on mouse CP to CARMIL binding**

Ikuko Fujiwara[1], Christopher Alexander[2], Kirsten Remmert[2], Grzegorz Piszcek[2], John Hammer[2] ([1]*NITech*, [2]*NHLBI, NIH*)

13:20～16:15　K 会場（107 教室）／Room K (107)
1K　分子モーター I / Molecular motor I

1K1320 **高度高塩菌ハロバクテリウムサリナラムの遊泳運動特性の解析**

Characterization of the swimming motility of halophilic archaea, *Halobacterium salinarum*

○木下 佳昭[1], 内田 就也[2], 中根 大介[1], 西坂 崇之[1]（[1]学習院大学 理・物理, [2]東北大学 理・物理）

Yoshiaki Kinosita[1], Nariya Uchida[2], Daisuke Nakane[1], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin University*, [2]*Department of Physics, Tohoku University*)

1K1335 **深海微生物の遊泳運動を高圧力下で観察する**

Direct observation of the swimming motility of deep-sea bacterium at high-pressure conditions

○西山 雅祥[1], 加藤 千明[2], 原田 慶恵[3]（[1]京大 白眉セ, [2]海洋研究開発機構, [3]京大 iCeMS）

Masayoshi Nishiyama[1], Chiaki Kato[2], Yoshie Harada[3] ([1]*The HAKUBI Center, Kyoto Univ.*, [2]*JAMSTEC*, [3]*WPI-iCeMS, Kyoto Univ.*)

1K1350 **クラミドモナス鞭毛の波形の切り替え制御因子**

A regulation factor responsible for switching waveform of Chlamydomonas flagella

○桐間 惇也[1], 白髪 美咲[1], 小嶋 寛明[2], 大岩 和弘[1,2]（[1]兵県大 院生命理学, [2](国) 情報通信研究機構）

Junya Kirima[1], Misaki Shiraga[1], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst, NICT*)

1K1405 **D32 のプロトン化で誘起されるべん毛モーター固定子 MotA/B の構造変化**

Structural change of the stator complex MotA/B on bacterial flagellar motor induced by protonation of D32

○西原 泰孝[1], 北尾 彰朗[2]（[1]東大院・総合文化, [2]東大・分生研）

Yasutaka Nishihara[1], Akio Kitao[2] ([1]*Univ. of Tokyo, CMSI*, [2]*Univ. of Tokyo, IMCB*)

休憩 14:20-14:25

1K1425 **Axonemal Dynein Light Chain-1 Locates at the Microtubule Binding Domain of the γ Heavy Chain**

Muneyoshi Ichikawa[1,2], Kei Saito[1], Haru-aki Yanagisawa[1], Toshiki Yagi[3], Ritsu Kamiya[4], Shin Yamaguchi[1], Junichiro Yajima[1], Yasuharu Kushida[5], Kentaro Nakano[5], Osamu Numata[5], Yoko Y. Toyoshima[1] ([1]*The Univ. of Tokyo*, [2]*McGill Univ.*, [3]*Pref. Univ. of Hiroshima*, [4]*Gakushuin Univ.*, [5]*Univ. of Tsukuba*)

1K1440 **Off-axis motion of yeast cytoplasmic dynein takes a biased random walk**

Mitsuhiro Sugawa[1], Shin Yamaguchi[1], Hiroaki Takagi[2], Keitaro Shibata[1,3], Yoko Y. Toyoshima[1], Junichiro Yajima[1] ([1]*Graduate School of Arts and Sciences, The University of Tokyo*, [2]*Department of Physics, Nara Medical University*, [3]*National Institute of Advanced Industrial Science and Technology*)

1K1455 **クライオ電子顕微鏡により明らかとなった微小管上を歩いている細胞質ダイニンの新規の構造と揺らぎ**

Direct observation of cytoplasmic dynein stepping on microtubules by cryo-EM reveals a novel hinge at stalk-stalkhead junction

○今井 洋[1,2], 島 知弘[3], 須藤 和夫[4], Walker Matthew L.[5], Knight Peter J.[2], 昆 隆英[6], Burgess Stan A.[2]（[1]中央大・理工, [2]英国リーズ大, [3]理研・QBiC, [4]早稲田大・理工, [5]MLW Consulting, [6]阪大・院理）

Hiroshi Imai[1,2], Tomohiro Shima[3], Kazuo Sutoh[4], Matthew L. Walker[5], Peter J. Knight[2], Takahide Kon[6], Stan A. Burgess[2] ([1]*Chuo Univ.*, [2]*Univ. of Leeds*, [3]*RIKEN QBiC*, [4]*Waseda Univ.*, [5]*MLW Consulting*, [6]*Grad. Sch. Sci., Osaka Univ.*)

1K1510 **神経細胞オルガネラ輸送におけるキネシンとダイニンの数の測定：揺らぎの定理の応用**
Measuring the numbers of kinesin and dynein on neuronal cargo transport using the fluctuation theorem
○林 久美子[1], 岡田 康志[2]（[1]東北大工, [2]理研QBiC）
Kumiko Hayashi[1], Yasushi Okada[2] ([1]*Sch. Eng., Tohoku Univ.*, [2]*QBiC, RIKEN*)

休憩 15:25-15:30

1K1530 **等方型 TRIFM とデフォーカスイメージングによる単一蛍光色素の角度と回転方向の検出**
Detection of 3-D orientation and rotation handedness of single fluorophore by isotropic TIRFM and defocused imaging
○藤村 章子[1], 伊藤 祐子[2], 足立 健吾[3], 池口 満徳[2], 西坂 崇之[1]（[1]学習院大学・理, [2]横浜市立大・生命医科学研究科, [3]早稲田大・理工）
Shoko Fujimura[1], Yuko Ito[2], Kengo Adachi[3], Mitsunori Ikeguchi[2], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin Univ.*, [2]*Medical Life Sci., Yokohama City Univ.*, [3]*Engin., Waseda Univ.*)

1K1545 **１分子の複数状態モニタリングにより明らかとなった F_1-ATPase におけるヌクレオチド周辺の局所環境と化学状態の相関**
Correlation between local environment around nucleotide and chemical state in F_1-ATPase revealed by single-molecule modes monitoring
○三上 渚, 西坂 崇之（学習院大・物理）
Nagisa Mikami, Takayuki Nishizaka (*Dept. phys., Gakushuin Univ.*)

1K1600 **Single Molecule Time Series Analysis of F1-ATPase to Unravel the Role of Bound-ATP Hydrolysis**
Chun-Biu Li, Tamiki Komatsuzaki (*RIES Hokkaido Univ.*)

13:20～16:15　L 会場（108 教室）／Room L (108)
1L　蛋白質：機能・計測・解析の方法論 / Proteins: Function, Measurement, Analysis

1L1320 **光センサータンパク質 TePixD の反応過程における過渡的揺らぎ**
Transient conformational fluctuation of TePixD during a reaction
○黒井 邦巧[1], 岡島 公司[2,3], 池内 昌彦[2], 徳富 哲[3], 寺嶋 正秀[4]（[1]分子研, [2]東大院・理, [3]大阪府大院・理, [4]京大院・理）
Kunisato Kuroi[1], Koji Okajima[2,3], Masahiko Ikeuchi[2], Satoru Tokutomi[3], Masahide Terazima[4] ([1]*Inst. for Mol. Sci.*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*Grad. Sch. Sci., Univ. Osaka Pref.*, [4]*Grad. Sch. Sci., Univ. Kyoto*)

1L1335 **ラマン分光法による高濃度抗体溶液のタンパク質間相互作用の研究**
The protein-protein interactions in highly concentrated antibody solution investigated by Raman spectroscopy
○太田 周志[1], 野口 慎太郎[1], 津本 浩平[2,3,4]（[1]堀場製作所 先行開発センター, [2]東大院工, [3]東大医科研, [4]東京大学創薬機構）
Chikashi Ota[1], Shintaro Noguchi[1], Kouhei Tsumoto[2,3,4] ([1]*Advanced R&D Center, Horiba, Ltd.*, [2]*School of Engineering, The University of Tokyo*, [3]*Institute of Medical Science, The University of Tokyo*, [4]*Drug Discovery Initiative, The University of Tokyo*)

1L1350 **Cys のチオレートと Trp のカチオン - π 相互作用による銅輸送タンパク質細胞外領域における一価銅安定化機構**
Cysteine- and Tryptophan-Based Copper(I) Stabilization in the Extracellular N-terminal Domain of Ctr4
○岡田 毬子, 三浦 隆史, 中林 孝和（東北大・院 薬）
Mariko Okada, Takashi Miura, Takakazu Nakabayashi (*Grad. Sch. Pharm. Sci. Tohoku Univ.*)

1L1405 **酸素の常磁性効果を利用したタンパク質の疎水性キャビティーの検出**
Molecular oxygen as a paramagnetic NMR probe of dynamic hydrophobic cavity in proteins
○北原 亮[1], 吉村 優一[2], Xue Mengjun[2], Mulder Frans A. A.[2]（[1]立命館大学薬学部, [2]オーフス大学化学）
Ryo Kitahara[1], Yuichi Yoshimura[2], Mengjun Xue[2], Frans A. A. Mulder[2] ([1]*College of Pharmaceutical Sciences, Ritsumeikan University*, [2]*Department of Chemistry and iNANO Center, University of Aarhus*)

休憩 14:20-14:25

1L1425 **一分子蛍光顕微鏡による p53 変異体の標的配列探索ダイナミクスの観察**
Observation of the Search Dynamics of p53 Mutants for the Target DNA Sequence by Single-molecule Fluorescence Microscopy
○伊藤 優志[1,2], 村田 崇人[1,2], 坂本 清志[1], 七谷 圭[3], 和田 健彦[1], 高橋 聡[1,2], 鎌形 清人[1,2]（[1]東北大・多元研, [2]東北大・院理学, [3]東北大・農学）
Yuji Itoh[1,2], Agato Murata[1,2], Seiji Sakamoto[1], Kei Nanatani[3], Takehiko Wada[1], Satoshi Takahashi[1,2], Kiyoto Kamagata[1,2] ([1]*IMRAM, Univ. Tohoku*, [2]*Grad. Sch. Sci., Univ. Tohoku*, [3]*Grad. Sch. Agr. Sci., Univ. Tohoku*)

1L1440 **DNA 結合蛋白質の単分子蛍光観察のための DNA 整列技術の開発**
Development of a new method for making the array of aligned DNAs, DNA garden, for the single-molecule fluorescence imaging
○五十嵐 千裕[1,2], 村田 崇人[1,2], 高橋 聡[1,2], 鎌形 清人[1,2]（[1]東北大多元研, [2]東北大院理）
Chihiro Igarashi[1,2], Agato Murata[1,2], Satoshi Takahashi[1,2], Kiyoto Kamagata[1,2] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. of Sci., Tohoku Univ.*)

1L1455 **X 線１分子追跡法・プローブ負荷試験による複合タンパク質・協同的運動の定量化**
Quantification of Cooperative Motions for Multi-subunit Proteins by Single Molecule Loading test with Diffracted X-ray Tracking
○関口 博史[1], 池崎 圭吾[2], 八木 直人[1], 佐々木 裕次[1,2] ([1]公益財団法人 高輝度光科学研究センター 利用研究促進部門, [2]東京大学大学院新領域創成科学研究科)
Hiroshi Sekiguchi[1], Keigo Ikezaki[2], Naoto Yagi[1], Yuji Sasaki[1,2] ([1]*JASRI/SPring-8*, [2]*Grad. School Frontier Sci., Univ. Tokyo*)

1L1510 **Hydrolysis of lipid droplets by artificially designed peptides**
Yoshihiro Iida, Atsuo Tamura (*Grad. Sci., Univ. Kobe*)

休憩 15:25-15:30

1L1530 **分子認識におけるメチル化の効果： 分子動力学計算による研究**
Effect of methylation on molecular recognition: A molecular dynamics study
○山下 雄史（東大先端研）
Takefumi Yamashita (*RCAST, Univ. Tokyo*)

1L1545 **Investigating kinetics of conformational change using molecular dynamics and milestoning**
Hiroshi Fujisaki[1], Ayori Mitsutake[2] ([1]*Nippon Medical School*, [2]*Keio Univ. Dep. Phys.*)

1L1600 **自由エネルギーパスサンプリングへの PaCS-MD の応用**
Application of PaCS-MD to Free Energy Path Sampling
○Tran Duy[1], 北尾 彰朗[1,2]（[1]東大 創域, [2]東大 分生研）
Duy Tran[1], Akio Kitao[1,2] ([1]*Grad. Frontier Sci., Univ. of Tokyo*, [2]*IMCB, Univ. of Tokyo*)

13:20〜16:15　M 会場（109 教室）／Room M (109)
1M　蛋白質：構造・構造機能相関 I / Proteins: Structure, Structure-function relationship I

1M1320 **Structural and functional comparison of hexahistidine tagged and untagged forms of small multidrug resistance protein, EmrE**
Shahzada Junaid S. Qazi, Raymond Chew, Denice C. Bay, Raymond J. Turner (*Biological Science Department, University of Calgary, Alberta, Canada*)

1M1335 **光駆動性ナトリウムイオンポンプによるナトリウムイオン輸送の構造基盤**
Structural basis for Na^+ transport mechanism by a light-driven Na^+ pump
○加藤 英明[1], 井上 圭一[2], 吉住 玲[2], 加藤 善隆[2], 大野 光[2], 今野 雅恵[2], 細島 頌子[3], 石塚 徹[3], Hoque Mohammad R.[3], 國友 博文[4], 伊藤 淳平[5], 吉澤 晋[6], 山下 恵太郎[7], 武本 瑞樹[4], 西澤 知宏[4], 谷口 怜哉[4], 木暮 一啓[6], Maturana Andres D.[5], 飯野 雄一[4], 八尾 寛[3], 石谷 隆一郎[4], 神取 秀樹[2], 濡木 理[4]（[1]スタンフォード大・医学, [2]名工大・院工, [3]東北大・院生命科, [4]東大・院理, [5]名大・院生命農, [6]東大・海洋研, [7]理研・播磨）
Hideaki Kato[1], Keiichi Inoue[2], Rei Yoshizumi[2], Yoshitaka Kato[2], Hikaru Ono[2], Masae Konno[2], Shoko Hososhima[3], Toru Ishizuka[3], Mohammad R. Hoque[3], Hirofumi Kunitomo[4], Jumpei Ito[5], Susumu Yoshizawa[6], Keitaro Yamashita[7], Mizuki Takemoto[4], Tomohiro Nishizawa[4], Reiya taniguchi[4], Kazuhiro Kogure[6], Andres D. Maturana[5], Yuichi Iino[4], Hiromu Yawo[3], Ryuichiro Ishitani[4], Hideki Kandori[2], Osamu Nureki[4] ([1]*Sch. of Med., Stanford Univ.*, [2]*Grad. Sch. of Engineering, Nagoya Inst. of Tech.*, [3]*Grad. Sch. of Life Sci., Tohoku Univ.*, [4]*Grad. Sch. of Sci., Univ. of Tokyo*, [5]*Grad. Sch. of Bioagri. Sci., Nagoya Univ.*, [6]*Atmos. and Ocean Res. Inst., Univ. of Tokyo*, [7]*Harima Inst., Riken SPring-8*)

1M1350 **コヒーレント X 線回折像から構造情報を抽出するための計算アルゴリズム**
Computational algorithms to extract structural information from X-ray coherent diffractions
○徳久 淳師[1], 宮下 治[1], タマ フロハンス[1,2]（[1]理研・計算科学研究機構, [2]名大・物理）
Atsushi Tokuhisa[1], Osamu Miyashita[1], Florence Tama[1,2] ([1]*RIKEN AICS*, [2]*Department of Physics, Nagoya University*)

1M1405 **X 線自由電子レーザーによって明らかにされた光化学系 II 複合体の 1.95Å 分解能での無損傷構造**
Radiation damage free structure of oxygen evolving photosytem II at 1.95Å resolution revealed by X-ray Free Electron Laser
○菅 倫寛[1], 秋田 総理[1], 平田 邦生[2], 上野 剛[2], 村上 博則[2], 中島 芳樹[1], 清水 哲哉[1], 山下 恵太郎[2], 山本 雅貴[2], 吾郷 日出夫[2], 沈 建仁[1]（[1]岡山大, [2]播磨理研）
Michihiro Suga[1], Fusamichi Akita[1], Kunio Hirata[2], Go Ueno[2], Hironori Murakami[2], Yoshiki Nakajima[1], Tetsuya Shimizu[1], Keitaro Yamashita[2], Masaki Yamamoto[2], Hideo Ago[2], Jian-Ren Shen[1] ([1]*Okayama Univ.*, [2]*Riken Harima*)

休憩 14:20-14:25

1M1425 **ファルネシル基結合型ヒトガレクチン 1 の構造**
Structure of Human Galectin-1 Binding Farnesyl Group
○平松 弘嗣, 山口 知美, 中林 孝和（東北大・院薬）
Hirotsugu Hiramatsu, Kazumi Yamaguchi, Takakazu Nakabayashi (*Grad. Sch. Pharm. Sci., Tohoku Univ.*)

1M1440 **ADP リボシル化酵素 C3 と RhoA 複合体の構造基盤**
Structural basis of ADP-ribosyltransferase C3 exoenzyme with RhoA complex
戸田 暁之, 鶴村 俊治, 吉田 徹, 津守 耶良, ○津下 英明（京都産業大学）
Akiyuki Toda, Toshiharu Tsurumura, Toru Yoshida, Yayoi Tsumori, **Hideaki Tsuge** (*Kyoto Sangyo University*)

1M1455 **ラン藻アナベナ由来 12 量体グルタミン合成酵素の結晶構造**
Dodecameric crystal structure of Anabaena Glutamine synthetase
○Toniti Waraphan[1], 吉田 徹[1], 鶴村 俊治[1], 津下 英明[1], 芦田 裕之[2], 高橋 香代[2], 澤 嘉弘[2]（[1]京都産業大学, [2]島根大学）
Waraphan Toniti[1], Toru Yoshida[1], Toshiharu Tsurumura[1], Hideaki Tsuge[1], Hiroyuki Ashida[2], Kayo Takahashi[2], Yoshihiro Sawa[2] ([1]*Kyoto Sangyo University*, [2]*Shimane University*)

1M1510 **GFP の A および B state における高分解能 X 線結晶構造解析**
X-ray crystallographic studies of GFP in the A and B states at high resolution
○高場 圭章, 竹田 一旗, 三木 邦夫（京大・院理）
Kiyofumi Takaba, Kazuki Takeda, Kunio Miki (*Grad. Sch. Sci., Kyoto Univ.*)

休憩 15:25-15:30

1M1530 **中性子結晶構造解析によるセルロース加水分解酵素中のプロトン伝達経路の観測**
Direct observation of proton pathway in cellulase by Neutron crystallography
○中村 彰彦[1], 石田 卓也[2], 日下 勝弘[3], 山田 太郎[3], 田中 伊知郎[3], 新村 信雄[3], 鮫島 雅弘[2], 五十嵐 圭日子[2]（[1]自然科学研究機構 岡崎統合バイオ, [2]東大院 農生科, [3]茨城大学）
Akihiko Nakamura[1], Takuya Ishida[2], Katuhiro Kusaka[3], Taro Yamada[3], Ichiro Tanaka[3], Nobuo Niimura[3], Masahiro Samejima[2], Kiyohiko Igarashi[2] ([1]*Okazaki Inst. for Integrative Bioscience*, [2]*Grad. Sch. Agr., Univ. Tokyo*, [3]*Univ. Ibaraki*)

1M1545 **バクテリオファージ P22 の 2 次元結晶化**
Two-dimensional crystallization of Bacteriophage P22
○吉村 英恭[1], エドワード イーサン[2], パターソン ダスティン[2], 内田 昌樹[2], マッコイ キンバリー[2], ロイコウドハリー ラジャーシ[2], シュワルツ ベンジャミン[2], ダグラス トレバー[2]（[1]明大理工, [2]インディアナ大学）
Hideyuki Yoshimura[1], Ethan Edwards[2], Dustin Patterson[2], Masaki Uchida[2], Kimberly McCoy[2], Rajarshi Roychoudhury[2], Benjamin Schwarz[2], Trevor Douglas[2] ([1]*Dep. Phys, Meiji Univ.*, [2]*Indiana Univ.*)

1M1600 **Blender による生物学的アニメーションの制作と共有**
Creating and shareing biological animations by Blender
○上野 豊（産業技術総合研究所）
Yutaka Ueno (*AIST Kansai*)

13:20〜14:20　N 会場（201 教室）／Room N (201)
1Na　バイオエンジニアリング / Bioengineering

1N1320 **生理的温度条件下での自律 DNA 計算に向けた DNA 生成反応システムの構築**
Construction of a DNA generation reaction system for autonomous DNA-based computing at a physiological temperature
○小宮 健, 董 克蘇, 竹中 健朗, 山村 雅幸（東工大・院総合理工）
Ken Komiya, Kesu Dong, Toshio Takenaka, Masayuki Yamamura (*Interdisci. Grad. Sch. of Sci. & Engi., Tokyo Tech.*)

1N1335 **細胞折紙：3 次元共培養システムの構築**
Cell origami technique for 3D cell co-culture system
○何 倩, 岡嶋 孝治, 繁富(栗林) 香織（北海道大学大学院情報科学研究科）
Qian He, Takaharu Okajima, Kaori Shigetomi(Kuribayashi) (*Graduate School of Information Science and Technology of Hokkaido University*)

1N1350 **マイクロピラーによる細胞核の力学的拘束は正常細胞の増殖を抑制するが腫瘍細胞には影響しない**
Mechanical trapping of the nucleus on microfabricated pillars inhibits the proliferation of normal cells but not tumor cells
○長山 和亮[1], 濱路 祐未[2], 佐藤 祐次[2], 松本 健郎[2]（[1]茨城大学工学部知能システム工学科, [2]名古屋工業大学工学部機械工学科）
Kazuaki Nagayama[1], Yumi Hamaji[2], Yuji Sato[2], Takeo Matsumoto[2] ([1]*Micro-Nano Biomechanics Laboratory, Department of Intelligent Systems Engineering, Ibaraki University*, [2]*Biomechanics Laboratory, Department of Mechanical Engineering, Nagoya Institute of Technology*)

1N1405 **血球細胞の流体力学的挙動の数値シミュレーション**
A numerical study on the fluid dynamics of blood cells
○Nix Stephanie[1], 今井 陽介[2], 石川 拓司[2,3]（[1]秋田県大・システム科学技術, [2]東北大・院工, [3]東北大・院医工）
Stephanie Nix[1], Yohsuke Imai[2], Takuji Ishikawa[2,3] ([1]*Fac. Sys. Sci. Tech., Akita Pref. Univ.*, [2]*Grad. Sch. Eng., Tohoku Univ.*, [3]*Grad. Sch. Biomed. Eng., Tohoku Univ.*)

14:25〜16:15　N 会場（201 教室）／Room N (201)
1Nb　膜蛋白質 I / Membrane proteins I

1N1425 **機械受容チャネルによるコリネ型細菌のグルタミン酸放出機構の解析**
Mechanism of glutamate export in Corynebacterium glutamicum through the mechanosensitive channel
○中山 義敬[1], 吉村 建二郎[2], 飯田 秀利[3], 川崎 寿[4], Kraemer Reinhard[5], Martinac Boris[1]（[1]ビクター・チャン心臓病研究所, [2]メリーランド大学, [3]東京学芸大, [4]東京電機大, [5]コロン大学）
Yoshitaka Nakayama[1], Kenjiro Yoshimura[2], Hidetoshi Iida[3], Hisashi Kawasaki[4], Reinhard Kraemer[5], Boris Martinac[1] ([1]*Victor Chang Cardiac Research Institute*, [2]*University of Maryland*, [3]*Tokyo Gakugei University*, [4]*Tokyo DENKI University*, [5]*University of Cologne*)

1N1440 **アンキリンリピートドメインと脂質の相互作用による TRPV1 チャネル活性の制御**
Channel activity regulatory mechanism of TRPV1 by the interaction of ankyrin repeat domain with phospholipids
○竹村 和浩[1], 末次 志郎[2], 北尾 彰朗[1]（[1]東大・分生研, [2]NAIST）
Kazuhiro Takemura[1], Shiro Suetsugu[2], Akio Kitao[1] ([1]*IMCB, Univ. Tokyo*, [2]*Grad. Sch. Biol. Sci., NAIST*)

1N1455 **真正細菌のポンプ型ロドプシンの機能転換およびそれらの光反応についての研究**
Functional conversion of eubacterial pump rhodopsins and the investigation of the photoreactions
○野村 祐梨香[1], 井上 圭一[1,2], 神取 秀樹[1]（[1]名工大・院工, [2]JSTさきがけ）
Yurika Nomura[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*PRESTO*)

1N1510 **膜輸送体の超高感度活性計測のための新しいマイクロデバイス**
Novel micro device for highly sensitive measurement of membrane transporter activities
○渡邉 力也[1,2], 曽我 直樹[1], 野地 博行[1]（[1]東京大学大学院工学系研究科応用化学専攻, [2]JST・さきがけ）
Rikiya Watanabe[1,2], Naoki Soga[1], Hiroyuki Noji[1] ([1]*Department of Applied Chemistry, The University of Tokyo*, [2]*PRESTO, JST*)

休憩 15:25-15:30

1N1530 **Mechanism of sodium/proton antiporter from transition path simulations**
Kei-ichi Okazaki, Judith Warnau, Gerhard Hummer (*Max Planck Institute of Biophysics*)

1N1545 **Molecular mechanisms of proton transfer in H^+-coupled MATE in outward facing form**
Wataru Nishima[1], Wataru Mizukami[2], Yoshiki Tanaka[3], Ryuichiro Ishitani[4], Osamu Nureki[4], Yuji Sugita[1] ([1]*RIKEN Theoretical Molecular Science Laboratory*, [2]*Department of Material Sciences, Faculty of Engineering Sciences, Kyushu University*, [3]*Laboratory of Membrane Molecular Biology, the Graduate School of Biological Sciences, Nara Institute of Science and Technology*, [4]*Department of Biophysics and Biochemistry, the University of Tokyo*)

1N1600 **Multiscale Dynamics of Flaviviridae Fusion Peptides: Membrane Interactions via Simulation and Experiment**
Jan Marzinek[1,2], Peter Bond[1,2], Chandra Verma[1,2,3] ([1]*National University of Singapore, Department of Biological Sciences, 14 Science Drive 4, Singapore 117543*, [2]*Bioinformatics Institute (A*STAR), 30 Biopolis Str., #07-01 Matrix, Singapore 138671*, [3]*School of Biological Sciences, Nanyang Technological University, 60 Nanyang Drive, Singapore 63755*)

13:20～16:15　O 会場（203 教室）／Room O (203)
1O　核酸結合蛋白質 / Nucleic acid binding proteins

1O1320 **転写因子 Ets1 天然変性領域のリン酸化による DNA 認識阻害メカニズムの検討**
Molecular Mechanism of Inhibition of Transcription Factor Ets1-DNA Binding, Induced by Phosphorylation on a Disordered Region
○笠原 浩太[1], 肥後 順一[1], 椎名 政昭[2], 緒方 一博[2], 中村 春木[1]（[1]阪大・蛋白研, [2]横浜市大院・医）
Kota Kasahara[1], Junichi Higo[1], Masaaki Shiina[2], Kazuhiro Ogata[2], Haruki Nakamura[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Med., Yokohama City Univ.*)

1O1335 **光活性化 bZIP モジュール Photozipper の DNA 結合性**
Evaluation of DNA-binding of a light-activatable bZIP module, Photozipper
○久冨 修, 矢部 悠生, 中谷 陽一（阪大・院理）
Osamu Hisatomi, Yuki Yabe, Yoichi Nakatani (*Grad. Sch. of Sci., Osaka Univ.*)

1O1350 **3 本鎖 DNA 結合蛋白質の 3 本鎖 DNA 認識機構**
Molecular mechanism of triplex DNA-binding proteins to recognize triplex DNA
○鳥越 秀峰, 木内 一樹, 間瀬 貴久江, 佐藤 憲大, 片山 拓馬（東京理科大学理学部応用化学科鳥越研究室）
Hidetaka Torigoe, Kazuki Kiuchi, Kikue Mase, Norihiro Sato, Takuma Katayama (*Department of Applied Chemistry, Faculty of Science, Tokyo University of Science*)

1O1405 **HU Binding Coupled Bending of Double Stranded DNA**
Cheng Tan, Tsuyoshi Terakawa, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

休憩 14:20-14:25

1O1425 **リバースジャイレースによる DNA の二重らせんをきつく巻きつける反応の解析**
Direct observation of DNA overwinding by reverse gyrase
○小川 泰策[1], 余語 克紀[2], 古池 晶[3], 須藤 和夫[1], 菊池 韶彦[4], 木下 一彦[1]（[1]早大・物理, [2]北里大院・医療系, [3]大阪医大・物理, [4]名大・医）
Taisaku Ogawa[1], Katsunori Yogo[2], Shou Furuike[3], Kazuo Sutoh[1], Akihiko Kikuchi[4], Kazuhiko Kinosita, Jr.[1] ([1]*Dept. Phys., Waseda Univ.*, [2]*Grad. Sch. Med. Sci., Kitazato Univ.*, [3]*Dept. Phys., Osaka Med. Coll.*, [4]*Grad. Sch. Med., Nagoya Univ.*)

1O1440 **Single-molecule measurement for sliding dynamics of tumor suppressor p53 on DNA**
Agato Murata[1,2], Yuji Itoh[1,2], Chihiro Igarashi[1,2], Dwiky Rendra Graha Subekti[1,3], Satoshi Takahashi[1], Kiyoto Kamagata[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad., Sch., Sci., Tohoku Univ.*, [3]*AMC, Fac., Sci., Tohoku Univ.*)

1O1455 **Direct visualization of *E.coli* SbcD enzymatic activity on single strand DNA by high-speed AFM**
Junyi Liang[1], Noriyuki Kodera[2,4], Hiromi Tanaka[2], Hiroki Konno[2,4], Toshio Ando[1,2,4] ([1]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [2]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [3]*PRESTO, JST*, [4]*CREST, JST*)

1O1510 **高速 AFM が捉えた大腸菌 MukB の構造動態**
Structural dynamics of *E. coil* MukB captured by HS-AFM
○米田 博紀[1], 矢野 晃一[2], 古寺 哲幸[3,4], 八木 健太[1], 仁木 宏典[2], 安藤 敏夫[1,3,5]（[1]金沢大・理工・数物科学, [2]遺伝研, [3]金沢大・理工・バイオ AFM, [4]JST・PRESTO, [5]JST・CREST）
Hironori Yoneda[1], Kouichi Yano[2], Noriyuki Kodera[3,4], Kenta Yagi[1], Hironori Niki[2], Toshio Ando[1,3,5] ([1]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [2]*Natl. Inst. of Genet.*, [3]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [4]*PRESTO, JST*, [5]*CREST, JST*)

休憩 15:25-15:30

1O1530 **Structural dynamics of tri-nucleosome by coarse-grained simulations: effects of histone acetylation**
Le Chang, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

1O1545 **粗視化分子シミュレーションによるクロマチン環境下における転写因子と ERK の拡散運動の研究**
The diffusion dynamics of transcription factors and ERK in chromatin environment studied by coarse-grained simulation
○金田 亮[1], 寺川 剛[1], 検崎 博生[2], 高田 彰二[1]（[1]京大・理, [2]理研・情基）
Ryo Kanada[1], Tsuyoshi Terakawa[1], Hiroo Kenzaki[2], Shoji Takada[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*ACCC, RIKEN*)

1O1600 **粗視化シミュレーションから見えてきたクロマチン凝縮ダイナミクスにおける H1 C 末端変性部位の重要性**
Disordered tail of the linker histone H1 in chromatin compaction dynamics studied by coarse-grained simulations
○白井 伸宙, 高田 彰二（京大・院理）
Nobu C. Shirai, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

13:20〜15:25　Q 会場（207 教室）／Room Q (207)
1Qa　生体膜・人工膜 I / Biological & Artificial membrane I

1Q1320 **固液界面における二成分系脂肪酸単分子膜の研究**
Study on structural formation of mixed fatty acid monolayers at liquid/solid interface
○日比野 政裕[1], 向井山 善嗣[2]（[1]室蘭工大・応理, [2]トヨタ自動車）
Masahiro Hibino[1], Yoshitsugu Mukaiyama[2] ([1]*Dept. Appl. Sci., Muroran Inst. Tech.*, [2]*Toyota Motor*)

1Q1335 **飽和リン脂質／コレステロール二成分混合膜系における相挙動の詳細解析**
Detailed phase behavior analysis in the saturated 1,2-diacylphosphatidylcholine/cholesterol binary monolayer system
○三好 翼, 加藤 知（関学大・院理工）
Tsubasa Miyoshi, Satoru Kato (*Grad. Sch. Sci & Tech., Univ. Kwansei Gakuin*)

1Q1350 **シミュレーションを用いた脂質二重膜の相転移の理論的研究**
A theoretical study of thermal phase transition of phospholipid bilayer
○緒方 浩二, 中村 振一郎（理化学研究所 イノベーション推進センター）
Koji Ogata, Shinichiro Nakamura (*RIKEN Innovation Center*)

1Q1405 **ER 膜タンパク質の膜貫通配列のリン脂質 flip-flop を促進する物理化学的性質の解析**
Physicochemical properties of membrane-spanning sequences in the ER proteins to promote phospholipid flip-flop
○中尾 裕之[1], 池田 恵介[2], 石濱 泰[1], 中野 実[2]（[1]京大院薬, [2]富山大院医薬）
Hiroyuki Nakao[1], Keisuke Ikeda[2], Yasushi Ishihama[1], Minoru Nakano[2] ([1]*Grad. Sch. Pharm. Sci., Kyoto Univ.*, [2]*Grad. Sch. Med. Pharm. Sci., Univ. Toyama*)

休憩 14:20-14:25

1Q1425 **シトクロム P450 基質薬剤クロルゾキサゾンのリン脂質 POPC 膜結合に対するコレステロールの阻害効果の X 線回折による研究**
X-ray diffraction studies of the effect of cholesterol on the binding of cytochrome P450 substrate drug chlorzoxazone to POPC bilayers
山田 安由美[1], 清水 伸隆[2], 引間 孝明[3], 高田 昌樹[3,4], 小林 俊秀[5], ○高橋 浩[1,3,5]（[1]群馬大院・理工, [2]高エネ研PF, [3]理研・播磨, [4]東北大・多元研, [5]理研）
Ayumi Yamada[1], Nobutaka Shimizu[2], Takaaki Hikima[3], Masaki Takata[3,4], Toshihide Kobayashi[5], **Hiroshi Takahashi**[1,3,5] ([1]*Grad. Sch. Sci & Tech., Gunma Univ.*, [2]*KEK-PF*, [3]*Harima Inst., Riken*, [4]*IMRAM, Tohoku Univ.*, [5]*Riken*)

1Q1440 **脂質ナノディスクを形成する膜活性ポリマー**
Membrane-active amphiphilic polymers for lipid bilayer nanodisc formation
○安原 主馬, 荒木田 臣, 菊池 純一（奈良先端大院物質）
Kazuma Yasuhara, Jin Arakida, Jun-ichi Kikuchi (*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*)

1Q1455 **ナノ粒子の細胞内取り込み機構：膜変形と粒子拡散**
Physical mechanism of cellular uptake of nanoparticles: membrane deformation and diffusion of adsorption particles
○執行 航希, 永井 健, 濵田 勉（北陸先端大 マテリアル）
Kazuki Shigyou, Ken Nagai, Tsutomu Hamada (*JAIST Material*)

1Q1510 **バナナ状タンパク質による膜チューブ形成**
Membrane tubulation induced by banana-shaped proteins
○野口 博司（東京大学 物性研究所）
Hiroshi Noguchi (*ISSP, Univ. Tokyo*)

15:30〜16:15　Q 会場（207 教室）／Room Q (207)
1Qb　非平衡・生体リズム / Nonequilibrium state & Biological rhythm

1Q1530　**電場に駆動される自発運動ゲル**
Self-propelled gel particles driven by electric field
○永井 健[1], 早川 雅之[2], 瀧ノ上 正浩[2,3]（[1]北陸先端科学技術大学院大学, [2]東工大院総理工, [3]JSTさきがけ）
Ken Nagai[1], Masayuki Hayakawa[2], Masahiro Takinoue[2,3] ([1]*JAIST*, [2]*Tokyo Tech.*, [3]*JST*)

1Q1545　**概日時計における周期の頑健性と位相の可塑性の互恵的関係**
Reciprocity between robustness of period and plasticity of phase in biological clocks
○畠山 哲央, 金子 邦彦（東京大学総合文化研究科）
Tetsuhiro S. Hatakeyama, Kunihiko Kaneko (*The University of Tokyo, Department of Basic Science*)

1Q1600　**フィードバックループのあるシグナル伝達におけるマックスウェルのデーモン**
Maxwell's demon in biochemical signal transduction with feedback loop
○伊藤 創祐[1], 沙川 貴大[2]（[1]東京工業大学, 物性物理学専攻, [2]東京大学, 工学系研究科）
Sosuke Ito[1], Takahiro Sagawa[2] ([1]*Department of Physics, Tokyo Institute of Technology*, [2]*Department of Applied Physics, the University of Tokyo*)

第 2 日目（9 月 14 日（月））／Day 2（Sep. 14 Mon.）

13:55〜16:00　B 会場（ファカルティーホール）／Room B (Faculty Hall)
2B　蛋白質：構造・構造機能相関 IV / Proteins: Structure, Structure-function relationship IV

2B1355　**アミロイド線維表面構造に対するヨウ素プローブの利用**
Utilizing iodine as a probe for surface structures of amyloid fibrils
○平松 貴人, ハ ソンミン, 増田 裕輝, 茶谷 絵理（神戸大・院理）
Takato Hiramatsu, Seongmin Ha, Yuki Masuda, Eri Chatani (*Grad. Sch. Sci., Kobe Univ.*)

2B1410　**テラヘルツ時間領域分光および誘電分光を用いたアミロイド線維水和水ダイナミクスの観測**
Hydration Water Dynamics of Amyloid Fibrils Studied by Terahertz Time-domain Spectroscopy and Dielectric Spectroscopy
○山本 直樹[1], 富永 圭介[1,2], 茶谷 絵理[1]（[1]神戸大学大学院理学研究科, [2]神戸大学分子フォトサイエンス研究センター）
Naoki Yamamoto[1], Keisuke Tominaga[1,2], Eri Chatani[1] ([1]*Graduate School of Science, Kobe University*, [2]*Molecular Photoscience Research Center, Kobe University*)

2B1425　**NMR による天然変性蛋白質の残存構造解析**
Residual structures in the intrinsically disordered proteins monitored by NMR
○西村 千秋（帝京平成大・薬）
Chiaki Nishimura (*Fac. Pharm. Sci., Teikyo Heisei Univ.*)

2B1440　**蛋白質高エネルギー構造パラダイムの役割**
The role of the "high-energy paradigm" of protein structure
○赤坂 一之（京都府立大・院生命環境科学）
Kazuyuki Akasaka (*Kyoto Pref. Univ., Grad. Sch., Bio. Envir. Sci.*)

2B1455　**シアノバクテリア由来アルカン合成関連酵素の機能解析及びバイオエネルギー生産への応用**
Functional analysis of the cyanobacterial enzyme for alkane biosynthesis and its application for bioenergy production
○工藤 恒[1], 名和 良太[2], 林 勇樹[1], 渡辺 麻衣[1], 池内 昌彦[1], 新井 宗仁[1,2]（[1]東大・総合文化・生命環境, [2]東大・教養・基礎科学科）
Hisashi Kudo[1], Ryota Nawa[2], Yuuki Hayashi[1], Mai Watanabe[1], Masahiko Ikeuchi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Pure & Applied Sci., Univ. Tokyo*)

休憩 15:10-15:15

2B1515　**Accelerated H-DROP: An SVM based Helical Domain linker pRedictor trained with OPtimized features**
Richa Tambi, Soichiro Ide, Ryosuke Suzuki, Teppei Ebina, Yutaka Kuroda (*Tokyo University of Agriculture and Technology*)

2B1530　**活性化システインを持つ蛋白質構造の理論設計**
Theoretical Design of Protein Scaffold Harboring an Activated Cysteine
○小杉 貴洋, Baker David（ワシントン大・生化）
Takahiro Kosugi, David Baker (*Dept. of Biochem., Univ. Washington*)

2B1545　**RE3Volutionary Computational Design of symmetric proteins**
Arnout Voet (*Yokohama, RIKEN CLST*)

8:45～10:00　C 会場（101 教室）／Room C (101)
2Ca　分子モーター II / Molecular motor II

2C0845　**金ナノロッドを用いたキネシン頭部構造変化の観察による協調的二足歩行の仕組みの解明**

Direct Observation of the Allosteric Conformational Change of Kinesin-1 using Gold Nanorod and its Implication for Head-head Coordination

○新谷 大和[1], 榎 佐和子[2], 野地 博行[2], 飯野 亮太[3], 富重 道雄[1]（[1]東京大学工学系研究科物理工学専攻, [2]東京大学工学系研究科応用化学専攻, [3]自然科学研究機構岡崎統合バイオサイエンスセンター分子科学研究所）

Yamato Niitani[1], Sawako Enoki[2], Hiroyuki Noji[2], Ryota Iino[3], Michio Tomishige[1] ([1]*Dept. Appl. Phys, Grad. Sch. Eng., Univ. Tokyo*, [2]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [3]*Okazaki Inst. Integ. BioSci., NINS*)

2C0900　**ミオシンの協調的首振りとアクチン滑り運動のゆらぎ**

Cooperative lever-arm swings of myosins and fluctuation of actin sliding

○近藤 洋太, 佐々木 一夫（東北大学）

Yota Kondo, Kazuo Sasaki (*Dept. Appl. Phys., Sch. Eng., Univ. Tohoku*)

2C0915　**プログラム可能な DNA バネにより明らかにされたミオシン VI の外力依存的なギアチェンジ機構**

Force-induced Gear-change Mechanism of Myosin VI Revealed with Programmed DNA Origami Spring

○岩城 光宏[1,2], ウィッカム シェリー[3,4,5], 池崎 圭吾[6], 柳田 敏雄[1,2,7], シー ウィリアム[3,4,5]（[1]理研・生命システム, [2]阪大院・生命機能, [3]ダナファーバー癌研, [4]ハーバード・医, [5]ハーバード・ヴィース研, [6]東大・新領域, [7]脳情報通信研）

Mitsuhiro Iwaki[1,2], Shelley Wickham[3,4,5], Keigo Ikezaki[6], Toshio Yanagida[1,2,7], William Shih[3,4,5] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*, [3]*Dana-Farber Cancer Inst.*, [4]*Harvard Med. Sch.*, [5]*Wyss Inst. Harvard Univ.*, [6]*Univ. of Tokyo*, [7]*CiNet*)

2C0930　**ダイニンとアクチン結合タンパク質を基に新しいモータータンパク質をデザインする**

Designing novel biomolecular motors based on dynein and actin-binding proteins

古田 茜, 大岩 和弘, 小嶋 寛明, ○古田 健也（国立研究開発法人 情報通信研究機構未来ICT研究所バイオICT研究室生体物性PJ）

Akane Furuta, Kazuhiro Oiwa, Hiroaki Kojima, **Ken'ya Furuta** (*Bio ICT Lab, NICT*)

2C0945　**マイコプラズマ・モービレの滑走装置に関わる新奇タンパク質**

Novel protein involving gliding machinery of *Mycoplasma mobile*

○浜口 祐, 田原 悠平, 松生 大輝, 宮田 真人（阪市大・院理）

Tasuku Hamaguchi, Yuhei O. Tahara, Daiki Matsuike, Makoto Miyata (*Grad. Sch. of Sci. Osaka City Univ.*)

13:55～16:15　C 会場（101 教室）／Room C (101)
2Cb　バイオイメージング II / Bioimaging II

2C1355　**テロメア繰り返し配列含有 RNA の生細胞内 1 分子イメージング**

Single molecule imaging of telomeric repeat-containinng RNA in living cells

○吉村 英哲, 山田 俊理, 瀬川 尋貴, 小澤 岳昌（東大・院理学系）

Hideaki Yoshimura, Toshimichi Yamada, Hiroki Segawa, Takeaki Ozawa (*Sch. Sci., Univ. Tokyo*)

2C1410　**Three dimensional trafficking of membrane protein PAR-1, labelled with quantum dot carried by endocytotic vesicles**

Seohyun Lee[1], Motoshi Kaya[1], Kohsuke Gonda[2], Hideo Higuchi[1] ([1]*Graduate School of Science, the University of Tokyo*, [2]*Graduate School of Medicine, Tohoku Univeristy*)

2C1425　**長時間１蛍光分子観察法の開発と応用：インテグリンの動的架橋が細胞接着を担う**

Development of long-term single fluorescent-molecule tracking revealed dynamic integrin crossbridging for cell adhesion

○角山 貴昭[1], 後藤 純理[1], 鈴木 健一[1,2], 藤原 敬宏[1], 楠見 明弘[1,3]（[1]京都大・物質-細胞統合システム拠点, [2]インド国立生命科学研究センターインド幹細胞・再生医学研究所, [3]京都大・再生研）

Taka-aki Tsunoyama[1], Junri Goto[1], Kenichi G.N. Suzuki[1,2], Takahiro K. Fujiwara[1], Akihiro Kusumi[1,3] ([1]*Inst. Integrated Cell-Material Sciences (WPI-iCeMS), Kyoto Univ.*, [2]*NCBS/inStem, India*, [3]*Inst. Frontier Medical Sciences, Kyoto Univ.*)

2C1440　**A fast- and positively photoswitchable fluorescent protein for ultralow-laser-power RESOLFT nanoscopy**

K Dhermendra Tiwari[1], Yoshiyuki Arai[1], Masahito Yamanaka[2], Tomoki Matsuda[1], Masakazu Agetsuma[1], Masahiro Nakano[1], Katsumasa Fujita[2], **Takeharu Nagai**[1] ([1]*ISIR, Osaka Univ.*, [2]*Dept. Appl. Phys., Osaka Univ.*)

休憩 14:55-15:15

2C1515　**高速光スイッチング蛍光タンパク質の蛍光偏光変調・励起角狭帯化照明による生体に優しい超解像イメージング**

Biocompatible superresolution imaging by polarization demodulation/excitation angle narrowing of fast photoswitching fluorescent proteins

○和沢 鉄一, 高内 大貴, Dhermendra Tiwari, 新井 由之, 松田 知己, 永井 健治（大阪大学 産業科学研究所）

Tetsuichi Wazawa, Hiroki Takauchi, Tiwari Dhermendra, Yoshiyuki Arai, Tomoki Matsuda, Takeharu Nagai (*ISIR, Osaka U*)

2C1530　**フェムト秒ファイバーレーザーで励起可能な高効率二光子蛍光分子**

Efficient Two-Photon Fluorescent Molecules Excitable by a Femtosecond Fiber Laser

○鈴木 康孝[1], 守友 博紀[1], 藤井 旺成[1], 白石 崇人[2]（[1]山口大学大学院医学系研究科, [2]山口大学理学部）

Yasutaka Suzuki[1], Hiroki Moritomo[1], Akinari Fuji[1], Takato Shiraishi[2] ([1]*Graduate School of Medicine, Yamaguchi University*, [2]*Faculty of Science*)

2C1545 **複数の光操作と組み合わせた膜電位メージングと薬剤スクリーニングの可能性を広げる発光指示薬の開発**

Luminescent indicator expands application for functional voltage imaging with multiple optical manipulation and drug screening

○稲垣 成矩[1], 松田 知己[1], 新井 由之[1], 神野 有香[2], 筒井 秀和[2,3], 岡村 康史[2]（[1]阪大・産研, [2]阪大・医学, [3]北陸先端大・マテリアル）

Shigenori Inagaki[1], Tomoki Matsuda[1], Yoshiyuki Arai[1], Yuka Jinno[2], Hidekazu Tsutsui[2,3], Yasushi Okamura[2] ([1]*ISIR., Univ. Osaka*, [2]*Grad. Sch. Med., Univ. Osaka*, [3]*Sch. Mat. Sci., JAIST*)

2C1600 **マルチカラー・リアルタイム生物発光イメージングのための３色の超高輝度発光タンパク質 Nano-lantern の開発**

Multicolor Nano-lanterns: the tricolored and super-brilliant luminescent proteins for multicolor, real-time bioluminescence imaging

○髙井 啓[1], 中野 雅裕[2], 齊藤 健太[2], 春野 玲弥[2], 渡邊 朋信[1,3], 大柳 達也[1], 神 隆[1], 岡田 康志[1], 永井 健治[1,2,3]（[1]理研QBiC, [2]阪大・産研, [3]JSTさきがけ）

Akira Takai[1], Masahiro Nakano[2], Kenta Saito[2], Remi Haruno[2], Tomonobu M. Watanabe[1,3], Tatsuya Ohyanagi[1], Takashi Jin[1], Yasushi Okada[1], Takeharu Nagai[1,2,3] ([1]*QBiC, RIKEN*, [2]*ISIR, Osaka Univ.*, [3]*PRESTO, JST*)

13:55〜16:15　D 会場（103 教室）／Room D (103)
2D　光生物：視覚・光受容 II / Photobiology: Vision & Photoreception II

2D1355 **FTIR 分光法を用いたイエロープロテインの発色団周囲の水素結合環境の解析**

Analysis of the hydrogen-bonding envirinment around the chromophore of photoactive yellow protein by FTIR spectroscopy

○岩田 達也[1,2], 神取 秀樹[1,2]（[1]名工大・院・工, [2]名工大・オプトバイオ）

Tatsuya Iwata[1,2], Hideki Kandori[1,2] ([1]*Dept. Eng., NITech*, [2]*OBtRC, NITech*)

2D1410 **MALDI-TOF-MS を用いた BLUF ドメインの同位体標識の解析**

Analysis of isotopic labeling of BLUF domain using MALDI-TOF-MS

○永井 貴士[1], 岩田 達也[1], 伊藤 奨太[1], 伊関 峰生[2], 渡辺 正勝[3], 北川 慎也[1], 神取 秀樹[1]（[1]名古屋工業大学, [2]東邦大学, [3]光産業創成大学院大学）

Takashi Nagai[1], Tatsuya Iwata[1], Shota Ito[1], Mineo Iseki[2], Masakatsu Watanabe[3], Shinya Kitagawa[1], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Toho University*, [3]*The Graduate School for the Creation of New Photonics Industries*)

2D1425 **青色光センサー蛋白質 BlrP1 の光反応ダイナミクス**

Photo-induced reaction dynamics of blue light sensory protein BlrP1

○柴田 耕生, 中曽根 祐介, 寺嶋 正秀（京大・院理化学）

Kousei Shibata, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci., Univ. Kyoto*)

2D1440 **光反応検出によって見えてくるフォトトロピンの多様性**

Diversity of phototropin studied from the viewpoint of photoreaction dynamics

○中曽根 祐介[1], 岡島 公司[2], 相原 悠介[1], 長谷 あきら[1], 徳富 哲[2], 寺嶋 正秀[1]（[1]京大・院理, [2]大阪府大・院理）

Yusuke Nakasone[1], Koji Okajima[2], Yusuke Aihara[1], Akira Nagatani[1], Satoru Tokutomi[2], Masahide Terazima[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Osaka Prefecture Univ.*)

2D1455 **遺伝子改変した LOV タンパク質中の FMN の光還元反応**

Photochemical reactions of FMN in the mutant LOV proteins

○上田 のぞみ, 小野 友紀子, 岩田 達也, 岩城 雅代, 神取 秀樹（名古屋工業大学大学院）

Nozomi Ueda, Yukiko Ono, Tatsuya Iwata, Masayo Iwaki, Hideki Kandori (*Nagoya Institute of Technology*)

休憩 15:10-15:15

2D1515 **Nonlabens dokdonesis DSW-6 由来微生物型ロドプシンの発現**

Expression of microbial rhodopsins in Nonlabens dokdonesis DSW-6

○橋本 優一, 阿部-吉住 玲, 神取 秀樹（名古屋工業大学）

Yuichi Hashimoto, Rei Abe-Yoshizumi, Hideki Kandori (*Nagoya Inst. Tech.*)

2D1530 **光駆動 Na^+-H^+KR2 におけるイオン取込み機構の研究**

Ion uptake mechanism of a light-driven Na^+-H^+ pump, KR2

○加藤 善隆[1], 井上 圭一[1,2], 吉住 玲[1], 神取 秀樹[1]（[1]名工大・院工, [2]PRESTO, JST）

Yoshitaka Kato[1], Keiichi Inoue[1,2], Rei Abe-Yoshizumi[1], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*PRESTO, JST*)

2D1545 **In situ 光照射固体 NMR によるバクテリオロドプシン Y185F 変異体の光反応経路の解明**

Photo-reaction pathways of bacteriorhodopsin Y185F mutant as revealed by in situ photo-irradiation solid-state NMR spectroscopy

大島 恭介[1], 重田 安里寿[1], 槙野 義輝[1], 川村 出[1], 沖津 貴志[2], 和田 昭盛[2], 辻 暁[3], 岩佐 達郎[4], ○内藤 晶[1] ([1]横浜国立大学 院工, [2]神戸薬大 生命有機, [3]兵庫県立大 院生命, [4]室蘭工大 院工)

Kyosuke Oshima[1], Arisu Shigeta[1], Yoshiteru Makino[1], Izuru Kawamura[1], Takashi Okitsu[2], Akimori Wada[2], Satoru Tuzi[3], Tatsuo Iwasa[4], **Akira Naito**[1] ([1]*Grad. Schl. Eng. Yokohama Natl. Univ.*, [2]*Dept. Org. Che. Life Sci. Kobe Pharm. Univ.*, [3]*Grad. Schl. Life Sci. Univ. Hyogo*, [4]*Grad. Schl. Eng. Muroran Ins. Tech.*)

2D1600 **プロトンドナーを持たない新たなプロトンポンプ型ロドプシン**

A new proton pump rhodopsin without a conserved proton donor

○鈴木 悠斗[1], 井上 圭一[1,2], Brown Lenonid S.[3], 神取 秀樹[1]（[1]名古屋工業大学大学院, [2]PRESTO JST, [3]University of Guelph）

Yuto Suzuki[1], Keiichi Inoue[1,2], Leonid S. Brown[3], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*PRESTO JST*, [3]*University of Guelph*)

13:55～16:15　E 会場（104 教室）／Room E (104)
2E　蛋白質：物性 / Proteins: Property

2E1355　天然変性タンパク質 HIV-1 Tat の pH 依存的な構造変化

pH-dependent conformational changes in the intrinsically disordered HIV-1 Tat protein

○椢原 朋子, 林 勇樹, 新井 宗仁（東大・総合文化・生命環境）

Tomoko Kunihara, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

2E1410　野生型及び病原性変異体 β2 ミクログロブリンの圧力変性反応の研究

Insights into the transition state of pressure-induced denaturation of β2-microglobulin and its pathogenic variants

○櫻井 一正[1], 豊増 明博[2], 前野 覚大[1], 橘 秀樹[2], 赤坂 一之[1]（[1]近畿大・先端研・高圧力蛋白研センター, [2]近畿大・生物理工）

Kazumasa Sakurai[1], Akihiro Toyomasu[2], Akihiro Maeno[1], Hideki Tachibana[2], Kazuyuki Akasaka[1] ([1]*HPPRC, Inst. Adv. Technol., Kindai Univ.*, [2]*BOST, Kindai Univ.*)

2E1425　ライン共焦点検顕微鏡を用いたマイクロ秒分解一分子 FRET 測定よる高速タンパク質折り畳みダイナミクスの追跡

Tracking microsecond single-molecule FRET dynamics on the fast protein folding by the line-confocal microscopy

○小井川 浩之[1], 新井 宗仁[2], 深澤 宏仁[3,4], 横田 浩章[4], 井出 徹[5], 高橋 聡[1]（[1]東北大・多元研, [2]東大・院総合文化, [3]浜松ホトニクス（株）, [4]光産業創成大学院大, [5]岡山大・院自然科学）

Hiroyuki Oikawa[1], Munehito Arai[2], Atsuhito Fukasawa[3,4], Hiroaki Yokota[4], Toru Ide[5], Satoshi Takahashi[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Dept. Life Sci., Univ. Tokyo*, [3]*Hamamatsu Photonics K.K.*, [4]*GPI*, [5]*Grad. Sch. Nat. Sci. and Tech., Okayama Univ.*)

2E1440　一分子蛍光分光法によって観測されたユビキチンの変性状態の不均一性

Conformational heterogeneity of denatured ubiquitin detected by single molecule fluorescence spectroscopy

○齊藤 雅嵩[1,2], Chen Eric H.-L.[3], Chen Po-Ting[3], Chen Rita P.-Y.[3], 鎌形 清人[1,2], 小井川 浩之[1,2], 高橋 聡[1,2]（[1]東北大多元研, [2]東北大院理, [3]IBC, Academia Sinica）

Masataka Saito[1,2], Eric H.-L. Chen[3], Po-Ting Chen[3], Rita P.-Y. Chen[3], Kiyoto Kamagata[1,2], Hiroyuki Oikawa[1,2], Satoshi Takahashi[1,2] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Sci., Tohoku Univ.*, [3]*IBC, Academia Sinica*)

2E1455　多数ペプチドから成る系の全原子分子動力学シミュレーションによるペプチド溶解性

Peptide solubility estimated by all-atom molecular dynamics simulation of multi-peptide systems concur with experimental values

○黒田 裕[1,2], 佐藤 雄士[2], 末永 敦[3], 泰地 真弘人[4]（[1]東京農工大学工学研究院生命機能科学部門, [2]東京農工大学大学院工学府生命工学専攻, [3]日本大学, [4]理化学研究所、生命システム研）

Yutaka Kuroda[1,2], Yuji Sato[2], Atsushi Suenaga[3], Makoto Taiji[4] ([1]*TUAT, Dept Biotech & Life Sci*, [2]*TUAT, Dept Biotech & Life Sci*, [3]*Nihon University*, [4]*Quantitative Biology Center, RIKEN*)

休憩 15:10-15:15

2E1515　出芽酵母を利用した SOD1 オリゴマー形成メカニズムの解明

A mechanism describing pathological oligomerization of SOD1 in a budding yeast model

○小野瀬 恭平, 西浦 由紘, 古川 良明（慶應大・理工・化学）

Kyohei Onose, Yuko Nishiura, Yoshiaki Furukawa (*Dept. Chem., Keio Univ.*)

2E1530　肝吸虫由来の銅シャペロンに着目した新たな SOD1 活性化メカニズムの解明

A new mechanism of SOD1 activation regulated by copper chaperone in Clonorchis sinensis

○福岡 真実[1], 長野 功[2], 古川 良明[1]（[1]慶應大・理工・化学, [2]岐阜大・医）

Mami Fukuoka[1], Isao Nagano[2], Yoshiaki Furukawa[1] ([1]*Dept. Chem., Keio Univ.*, [2]*Gifu Univ. Sch. Med.*)

2E1545　ペプチドを利用したミスフォールド型 SOD1 タンパク質の新たな検出手法

A new peptide-based method to detect misfolded SOD1 proteins

○長澤 健一, 野村 尚生, 古川 良明（慶應大・理工・化学）

Kenichi Nagasawa, Takao Nomura, Yoshiaki Furukawa (*Dept. Chem., Keio Univ.*)

2E1600　スーパーオキシドディスムターゼにおける新たな金属イオン獲得経路

A new intramolecular route to acquire metal ions in superoxide dismutase

○小久保 鉄平, 櫻井 靖之, 古川 良明（慶應大・理工・化学）

Teppei Kokubo, Yasuyuki Sakurai, Yoshiaki Furukawa (*Dept. Chem., Keio Univ.*)

13:55～15:10　J 会場（105 教室）／Room J (105)
2Ja　細胞生物的課題 II / Cell biology II

2J1355　超解像光学顕微鏡で観察した、収縮環におけるアクチンフィラメントとミオシンの配置

Arrangement of actin filaments and myosins in contractile ring, revealed with super-resolution microscopy

○加藤 薫[1], 上条 桂樹[2], 高橋 正行[3], 細谷 浩史[1,4]（[1]産総研バイオメディカル, [2]東北大・医・人体構造, [3]北大・理・化学, [4]学習院・理・生命科学）

Kaoru Katoh[1], Keijyu Kamijo[2], Masayuki Takahashi[3], Hiroshi Hosoya[1,4] ([1]*Biomed. Res. Inst, AIST*, [2]*Dept. Anat. & Anthropol, Sch. Med., Tohoku Univ.*, [3]*Dept. Chem., Grad. Sch. Sci., Hokkaido Univ.*, [4]*Dept Biol Sci., Fac Sci, Gakushuin Univ.*)

2J1410 **走化性タンパク質の発現量が大腸菌の走化性シグナル伝達系に与える影響**

Effect of the expression level of the chemotaxis proteins to the kinetic property of the signal processing of *Escherichia coli*

○佐川 貴志[1], 田中 裕人[1], 曽和 義幸[2], 川岸 郁朗[2], 小嶋 寛明[1]（[1]情報通信研究機構, [2]法政大 生命機能）

Takashi Sagawa[1], Hiroto Tanaka[1], Yoshiyuki Sowa[2], Ikuro Kawagishi[2], Hiroaki Kojima[1] ([1]*NICT*, [2]*Dept. of Frontier Biosci., Hosei Univ.*)

2J1425 **滑走するバクテリアの自己集合により形成される巨大渦の一方向性回転**

Directional rotation of large-scale vortex made of self-assembly of gliding bacteria

○小高 祥子, 中根 大介, 西坂 崇之（学習院大学 理学部 物理学科）

Showko Odaka, Daisuke Nakane, Takayuki Nishizaka (*Dept. Phys., Gakushuin Univ.*)

2J1440 **ゲル上での細胞集合体形成における力学モデルの検討**

Mechanical Study of Formation of Cell Aggregates on Hydrogel

○下川 裕子[1], 小池 博之[3], 松崎 賢寿[1], 江野村 允宏[2], 木村 昌樹[2], 中林 誠一郎[1], 中内 啓光[3], 谷口 英樹[2], 武部 貴則[2,3], 吉川 洋史[1]（[1]埼玉大・院理工, [2]横浜市大・院 医, [3]スタンフォード大・医）

Yuko Shimokawa[1], Hiroyuki Koike[3], Takahisa Matsuzaki[1], Masahiro Enomura[2], Masaki Kimura[2], Seiichiro Nakabayashi[1], Hiromitsu Nakauchi[3], Hideki Taniguchi[2], Takanori Takebe[2,3], Hiroshi Yoshikawa[1] ([1]*Dept. Chem., Saitama Univ.*, [2]*Dept. Regen. Med., YCU*, [3]*ISCBRM, Dept. Med., Stanford Univ.*)

2J1455 **マウス気管繊毛の非対称運動メカニズム ー軸糸の変形の視点からー**

Mechanism of asymmetric beating of mouse tracheal cilia - *from the perspective of the axoneme bending* -

○加藤 孝信[1], 池上 浩司[2], 内田 就也[3], 岩瀬 寿仁[4], 政池 知子[4,5], 瀬藤 光利[2], 西坂 崇之[1]（[1]学習院大・理・物理, [2]浜松医大・解剖学細胞生物学, [3]東北大・理・物理, [4]東理大・理工・応用生物, [5]科学技術振興機構・さきがけ）

Takanobu A Katoh[1], Koji Ikegami[2], Nariya Uchida[3], Toshihito Iwase[4], Tomoko Masaike[4,5], Mitsutoshi Setou[2], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin Univ.*, [2]*Dept. Cell Biol. and Anat., Hamamatsu Univ. Sch. Med.*, [3]*Dept. Phys., Tohoku Univ.*, [4]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [5]*PRESTO, JST*)

15:15～16:15　J 会場（105 教室）／Room J (105)
2Jb　ヘム蛋白質 / Heme proteins

2J1515 **チトクローム c 酸化酵素における酸素還元反応とプロトンポンプ共役機構**

The coupling mechanisms of oxygen reduction reaction and proton pumping in cytochrome c oxidase

○中島 聡, 西口 達人, 李 辰, 伊藤-新澤 恭子, 吉川 信也, 小倉 尚志（兵県大・院生命理学）

Satoru Nakashima, Tatsuhito Nishiguchi, Chen Li, Kyoko Shinzawa-Itoh, Shinya Yoshikawa, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

2J1530 **蛍光減少率測定によるヘムオキシゲナーゼ変異体ー電子供与蛋白相互作用の解析**

Fluorescence decay rates reveal the unique interactions between heme oxygenase mutants and its electron-donor protein

○三宅 倫生, 右田 たい子（山口大・農）

Norio Miyake, Taiko Migita (*Fac. of Agri., Dep. of Biol. Chem., Yamaguchi Univ.*)

2J1545 **亜硝酸還元酵素と一酸化窒素還元酵素の複合体形成による効率的な一酸化窒素分解機構**

Formation of Complex between Nitrite Reductase and Nitric Oxide Reductase for Rapid NO Elimination

○當舎 武彦[1], 寺坂 瑛里奈[1,2], 松本 喜慎[1,2], 杉本 宏[1], 城 宜嗣[1,2]（[1]理研・SPring-8, [2]兵県大・院生命理）

Takehiko Tosha[1], Erina Terasaka[1,2], Kimi Matsumoto[1,2], Hiroshi Sugimoto[1], Yoshitsugu Shiro[1,2] ([1]*RIKEN SPring-8*, [2]*Grad. Sch. Sci., Univ. Hyogo*)

2J1600 **マイクロ流路フローフラッシュ赤外吸収分光法の開発と一酸化窒素還元酵素の反応中間体解析への応用**

Development of Micro-channel Flow-flash Infrared Absorption Spectroscopy and its Application to Intermediate of Nitric Oxide Reductase

○木村 哲就[1], 石井 頌子[1,2], 當舎 武彦[1], 城 宜嗣[1,2], 久保 稔[1,3]（[1]理研, [2]兵県大・院生命理学, [3]JST・さきがけ）

Tetsunari Kimura[1], Shoko Ishii[1,2], Takehiko Tosha[1], Yoshitsugu Shiro[1,2], Minoru Kubo[1,3] ([1]*SPring-8 Cent., RIKEN*, [2]*Grad. Sch. Sci., Univ. Hyogo*, [3]*PRESTO, JST*)

13:55～16:15　K 会場（107 教室）／Room K (107)
2K　蛋白質：構造・構造機能相関 III / Proteins: Structure, Structure-function relationship III

2K1355 **タンパク質フォールディングの動的機構の理論的研究**

Theoretical study of the dynamic protein folding/unfolding transition mechanisms

○森 俊文, 斉藤 真司（分子研）

Toshifumi Mori, Shinji Saito (*IMS*)

2K1410 **Mechanisms of selection of channel for substrate transport in GatCAB**

Jiyoung Kang, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

2K1425 **Characterizing the gating of Skp: a periplasmic chaperone**

Daniel Holdbrook[1], Bjorn Burmann[2], Sebastian Hiller[2], Peter Bond[1] ([1]*Bioinformatics Institute, A*STAR*, [2]*Biozentrum, University of Basel, Basel, Switzerland*)

2K1440 **トレオニン合成酵素における生成物支援触媒機構についての理論的解明**
Theoretical elucidation on the molecular mechanism of product assisted catalysis of threonine synthase
○庄司 光男[1], 氏家 謙[1], 栢沼 愛[1], 重田 育照[1], 村川 武志[2], 林 秀行[2]（[1]筑波大, [2]大阪医大）
Mitsuo Shoji[1], Yuzuru Ujiie[1], Megumi Kayanuma[1], Yasuteru Shigeta[1], Takeshi Murakawa[2], Hideyuki Hayashi[2] ([1]*University of Tsukuba*, [2]*Osaka Medical College*)

2K1455 **Reaction mechanism of glycinamide ribonucleotide synthetase: Free-energy profile of the formation of an acylphosphate intermediate**
Norifumi Yamamoto[1], Gen-ichi Sampei[2], Gota Kawai[1] ([1]*Chiba Tech*, [2]*Univ Electro Comm*)

休憩 15:10-15:15

2K1515 **Exploring N-glycan conformers: assessment of force fields and enhanced sampling algorithms**
Raimondas Galvelis, Suyong Re, Yuji Sugita (*RIKEN TMSL*)

2K1530 **Oligomerization of Aβ fragments by the Hamiltonian replica-permutation method**
Satoru Itoh[1,2], Hisashi Okumura[1,2] ([1]*IMS*, [2]*Sokendai*)

2K1545 **拡張アンサンブル分子動力学シミュレーションを用いた電子顕微鏡データからの構造精密化手法**
New Approach for Cryo-EM Data Flexible Fitting Using Generalized Ensemble Molecular Dynamics Simulation
○宮下 治[1], 小林 千草[1], 森 貴治[1,2], 杉田 有治[1,2], タマ フロハンス[1,3]（[1]理研・計算科学, [2]理研・杉田理論分子化学, [3]名古屋大学・物理）
Osamu Miyashita[1], Chigusa Kobayashi[1], Takaharu Mori[1,2], Yuji Sugita[1,2], Florence Tama[1,3] ([1]*RIKEN AICS*, [2]*RIKEN TMSL*, [3]*Nagoya Univ. Phys.*)

2K1600 **単原子イオン周囲のハイパーモバイル水は多体効果に起因する**
Hypermobile Water around Monoatomic Ions Is Derived from Many-Body Effect
○最上 譲二[1], 松林 伸幸[2], 鈴木 誠[1]（[1]東北大・院工, [2]大阪大・院基礎工）
George Mogami[1], Nobuyuki Matubayasi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Eng. Sci., Osaka Univ.*)

13:55～15:10　L 会場（108 教室）／Room L (108)
2L　生命情報科学 / Bioinfomatics

2L1355 **Novel algorithm for identification of gene clusters in whole genome DNA sequences by combining rigorous and heuristic schemes**
Yuhya Takahashi, Jiyoung Kang, Masaru Tateno (*Grad. Sch. Life Sci., Univ. Hyogo*)

2L1410 **DELTA-FORTE: a profile-profile comparison method enhanced by curated database**
Toshiyuki Oda[1], Kazunori Yamada[1,2], Kentaro Tomii[1] ([1]*Biotech. Res. Inst. for Drug Discovery, AIST*, [2]*Grad. Sch. Info. Sci., Tohoku Univ.*)

2L1425 **A Clustering Approach to Visualize the Sequence-Structure-Function Relationship of Protein Enzymes**
Te-Lun Mai, Geng-Ming Hu, Chi-Ming Chen (*Department of Physics, National Taiwan Normal University*)

2L1440 **ホモ二量体タンパク質の立体構造変化の構造的、機能的特徴**
Structural and functional characterization of structural changes in homodimeric proteins
○雨宮 崇之, 堀井 達哉, 小池 亮太郎, 太田 元規（名大院・情報）
Takayuki Amemiya, Tatsuya Horii, Ryotaro Koike, Motonori Ota (*Grad. Schl. of Info. Sci., Nagoya Univ.*)

2L1455 **ハブタンパク質のいるところ**
Where the hub proteins are
○太田 元規[1], 権蛇 日出輝[1], 小池 亮太郎[1], 福地 佐斗志[2]（[1]名大・情科, [2]前工大・工）
Motonori Ota[1], Hideki Gonjya[1], Ryotaro Koike[1], Satoshi Fukuchi[2] ([1]*Grad. Sch. Info. Sci., Nagoya U.*, [2]*Fac. Eng., M.I.T.*)

13:55～15:10　M 会場（109 教室）／Room M (109)
2Ma　筋肉 / Muscle

2M1355 **細いフィラメント上のトロポミオシンの位置を ESR 距離測定により探索する**
Searching for tropomyosin position in the thin filament by distance measurements using spin-labeling dipolar EPR spectroscopy
○荒田 敏昭[1], 植田 啓介[2], 辻元 由起[1], 三木 正雄[3]（[1]阪大・院理・生物科学, [2]理研・横浜, [3]福井大学）
Toshiaki Arata[1], Keisuke Ueda[2], Yoshiki Tsujimoto[1], Masao Miki[3] ([1]*Dept. Biol., Sci. Grad. Sch., Osaka Univ.*, [2]*RIKEN-Yokohama*, [3]*Univ. Fukui*)

2M1410 **骨格筋ミオシン分子間の力発生が同調する仕組み**
Mechanism of cooperative force generation between skeletal myosins
○茅 元司[1], 鷲尾 巧[2], 久田 俊明[2], 樋口 秀男[1]（[1]東京大学 院理物理, [2]東京大学 大学院新領域創成研究科）
Motoshi Kaya[1], Takumi Washio[2], Toshiaki Hisada[2], Hideo Higuchi[1] ([1]*Graduate School of Science, University of Tokyo*, [2]*Graduate School of Frontier Science, University of Tokyo*)

2M1425 **蛋白質ダイナミクスから観た筋収縮調節機構：筋肉の細いフィラメントの中性子散乱による研究**
A view of the regulatory mechanism of muscle contraction from protein dynamics: a neutron scattering study of muscle thin filaments
○藤原 悟[1], 松尾 龍人[1], 山田 武[2], 柴田 薫[3]（[1]日本原子力研究開発機構量子ビーム応用研究センター, [2]総合科学研究機構, [3]J-PARCセンター）
Satoru Fujiwara[1], Tatsuhito Matsuo[1], Takeshi Yamada[2], Kaoru Shibata[3] ([1]*QuBS, JAEA*, [2]*CROSS-Tokai*, [3]*J-PARC Center*)

2M1440 **心筋の昇温誘起高速サルコメア振動**

Hyperthermal Sarcomeric Oscillations in cardiac muscle

○新谷 正嶺[1,2], 樋口 秀男[1], 福田 紀男[3], 石渡 信一[4]（[1]東大・物理, [2]学振PD, [3]慈恵医大・細生理, [4]早大・物理）

Seine Shintani[1,2], Hideo Higuchi[1], Norio Fukuda[3], Shin'ichi Ishiwata[4] ([1]*Dept. Phys., Univ. Tokyo*, [2]*JSPS Research Fellow*, [3]*Dept. Cell Phys., Sch. Med., Univ. Jikei*, [4]*Dept. Phys., Univ. Waseda*)

2M1455 **Disorder profile of nebulin encodes a vernier-like position sensor**

Ming-Chya Wu[1,2], Jeffrey G. Forbes[3], Kuan Wang[2,4] ([1]*RCADA, NCU, Taiwan*, [2]*IP, AS, Taiwan*, [3]*TWIC, Inc, USA*, [4]*IBC, AS, Taiwan*)

15:15～16:00　M 会場（109 教室）／Room M (109)
2Mb　神経科学 / Neuroscience

2M1515 **電気刺激により引き起こされるグラスキャットフィッシュの電気受容求心性線維の発火の非線形特性**

Nonlinear characteristics of electrosensory afferent nerve impulses elicited by sinusoidal electric stimulation in glass catfish

○足立 侑駿, 立野 勝巳（九州工業大学）

Yu Adachi, Katsumi Tateno (*Kyushu Institute of Technology*)

2M1530 **膜電位感受性色素による神経回路機能のアッセイ系の構築ー海馬スライスとビスフェノール A**

Neural circuit functional assay with voltage-sensitive dye imaging in hippocampal slices; effect of maternal bisphenol A

○冨永 貴志[1], 冨永 洋子[1], 五十嵐 勝秀[2,3], 大塚 まき[2,3], 古川 佑介[2], 菅野 純[2], 種村 健太郎[4]（[1]徳島文理大・神経研, [2]衛生研・毒性部, [3]星薬科大・L-StaR, [4]東北大院・農・動物生殖科学）

Takashi Tominaga[1], Yoko Tominaga[1], Katsuhide Igarashi[2,3], Maky Otsuka I[2,3], Yusuke Furukawa[2], Jun Kanno[2], Kentaro Tanemura[4] ([1]*Inst. Neurosci., Tokushima Bunri Univ.*, [2]*Div Cellular & Molecular Toxicol, NIHS*, [3]*L-StaR, Hoshi Univ. Sch. Pharmacy Pharmaceutical Sci.*, [4]*Lab Animal Reproduction, Grad Sch Agr Sci.*)

2M1545 **フェムト秒レーザー光刺激による神経細胞ネットワークの機能解析**

Functional analysis of living neuronal networks with a femtosecond laser-induced stimulation

○中川 裕太[1,2], 工藤 卓[2], 田口 隆久[3], 細川 千絵[1]（[1]産総研・バイオメディカル, [2]関西学院大院・理工, [3]情通機構・脳情報）

Yuta Nakagawa[1,2], Suguru N. Kudoh[2], Takahisa Taguchi[3], Chie Hosokawa[1] ([1]*Biomed. Res. Inst., AIST*, [2]*Grad. Sch. Sci. & Tech., Kwansei Gakuin Univ.*, [3]*CiNet, NICT*)

13:55～15:10　N 会場（201 教室）／Room N (201)
2Na　膜蛋白質 II / Membrane proteins II

2N1355 **補酵素置換による微生物外膜シトクロムの電子伝達速度制御**

Rate Regulation of *in vivo* Extracellular Electron Transport by Replacing Flavin Cofactors in Outer-membrane *C*-type Cytochromes

○徳納 吉秀, 岡本 章玄, 橋本 和仁（東京大学工学系研究科応用化学専攻）

Yoshihide Tokunou, Akihiro Okamoto, Kazuhito Hashimoto (*Department of applied chemistry, Univ. Tokyo*)

2N1410 **Uniquely Small Outer Membrane Cytochrome-c as a Possible Electron Carrier for Direct Electron Uptake in a Sulfate Reducing Bacterium**

Xiao Deng, Akihiro Okamoto, Kazuhito Hashimoto (*Grad. Sch. Eng., Univ. Tokyo*)

2N1425 **酸素センサータンパク質 Aer のシグナル伝達機構の解明**

Elucidation of signal transduction mechanism of Aer

○岡 頼良, 岩田 達也, 岩城 雅代, 神取 秀樹（名古屋工業大学）

Yoriyoshi Oka, Tatsuya Iwata, Masayo Iwaki, Hideki Kandori (*Nagoya Institute of Technology*)

2N1440 **一分子 FRET 観察によるセンサリーロドプシン-トランスデューサー複合体の構造変化の研究**

Single-molecule FRET studies on the light-induced structural changes of the sensory rhodopsin-transducer signaling complex

○西村 嶺[1], 井上 圭一[1,2], 谷ヶ崎 仁[3], 川本 健一[1], 須藤 雄気[4], 神取 秀樹[1]（[1]名古屋工業大学, [2]JST さきがけ, [3]名古屋大学, [4]岡山大学）

Ryo Nishimura[1], Keiichi Inoue[1,2], Jin Yagasaki[3], Kenichi Kawamoto[1], Yuki Sudo[4], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*JST PRESTO*, [3]*Nagoya University*, [4]*Okayama Uiversity*)

2N1455 **銅輸送 P 型 ATPase における ATP 加水分解中の イオン結合構造の ESR 解析**

ESR Spectroscopy on Metal binding Sites in Cu^{2+} -Transporting Protein during ATP Hydrolysis

○安田 哲[1], 堀本 拓也[1], 大門 大朗[1], 植田 恭広[1], 桑原 直之[3], 荒田 敏昭[1,2]（[1]阪大・理学・生物, [2]阪大・理学・先端強磁場センター, [3]高エネ研PF）

Satoshi Yasuda[1], Takuya Horimoto[1], Hiroaki Daimon[1], Yasuhiro Ueda[1], Naoyuki Kuwabara[3], Toshiaki Arata[1,2] ([1]*Dept. Biol. Sci., Osaka Univ.*, [2]*Ctr. High Mag, Field Sci., Grad. Sch. Sci., Osaka Univ.*, [3]*Photon Factory, KEK*)

15:15～16:15　N 会場（201 教室）／Room N (201)
2Nb　発生・分化 / Development & Differentiation

2N1515　**ES 細胞の分化における単一ヌクレオソームのイメージング**

Single nucleosome imaging in the differentiation of embryonic stem cells

○端保 舞[1,2], 野崎 慎[1,3], 田村 佐知子[1,2], 前島 一博[1,2]（[1]遺伝研, [2]総研大・生命科学, [3]慶応大・先端生命科学）

Mai Tambo[1,2], Tadasu Nozaki[1,3], Sachiko Tamura[1,2], Kazuhiro Maeshima[1,2] ([1]*Natl. Inst. of Genet.*, [2]*Sch. of Life Sci. The Grad. Univ. for Adv. Studies*, [3]*Inst. for Adv. Biosci., Keio Univ.*)

2N1530　**幹細胞の確率的分化ダイナミクスのつくる空間パターン**

Spatial Pattern in Stochastic Dynamics of Stem Cell Differentiation

○山口 裕樹[1], 川口 喬吾[2], 沙川 貴大[3]（[1]東大総合文化, [2]ハーバード医大, [3]東大工）

Hiroki Yamaguchi[1], Kyogo Kawaguchi[2], Takahiro Sagawa[3] ([1]*Dept. Bas. Sci., Univ. Tokyo*, [2]*Dept. Sys. Biol., Harvard Med. Sch.*, [3]*Dept. Appl. Phys., Univ. Tokyo*)

2N1545　**成長する上皮管の径サイズを維持する動的な細胞挙動について**

On Dynamic Cellular Behaviors Realizing Stable Radial Size in Developing Epithelial Tubule

○平島 剛志, 安達 泰治（京都大学再生医科学研究所）

Tsuyoshi Hirashima, Taiji Adachi (*Inst Front Med Sci, Kyoto Univ*)

2N1600　**枯葉に擬態した蝶の模様の進化**

Evolution of leaf mimicry in butterfly wing patterns

○鈴木 誉保, 冨田 秀一郎, 瀬筒 秀樹（国立研究開発法人 農業生物資源研究所 遺伝子組換えカイコユニット）

Takao K Suzuki, Shuichiro Tomita, Hideki Sezutsu (*National Institute of Agrobiological Sciences*)

13:55～15:10　O 会場（203 教室）／Room O (203)
2Oa　光生物：光合成 / Photobiology: Photosynthesis

2O1355　**PELDOR study on the high-affinity Mn(II) site of photoactivation of photosystem II**

Mizue Asada, Hiroyuki Mino (*Grad. Sch. Sci., Nagoya Univ.*)

2O1410　**時間分解 EPR 法によるホウレン草の光合成光化学系 II 反応中心における初期電荷分離構造**

Time-Resolved EPR Study on Geometry and Dynamics of the Primary Charge-Separated State in the Photosystem II Reaction Center of Spinach

○小堀 康博[1], 長谷川 将司[1], 立川 貴士[1], 近藤 徹[2], 長島 宏樹[3], 酒井 貴弘[3], 三野 広幸[3]（[1]神戸大院理, [2]東工大院理工, [3]名古屋大院理）

Yasuhiro Kobori[1], Masashi Hasegawa[1], Takashi Tachikawa[1], Toru Kondo[2], Hiroki Nagashima[3], Takahiro Sakai[3], Hiroyuki Mino[3] ([1]*Graduate School of Science, Kobe University*, [2]*Tokyo Institute of Technology*, [3]*Graduate School of Science, Nagoya University*)

2O1425　**FTIR spectroelectrochemical measurement of the redox potential of the secondary quinone electron acceptor Q_B in photosystem II**

Yuki Kato, Ryo Nagao, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

2O1440　**光化学系 II におけるチロシン Y_D からのプロトン放出：FTIR 法による検出**

FTIR detection of proton release from the redox-active tyrosine Y_D in photosystem II

○中村 伸, 野口 巧（名古屋大院・理）

Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

2O1455　**光合成水分解反応における Yz 周辺の水素結合ネットワークの役割**

FTIR study on the role of hydrogen bond network around Yz during photosynthetic water oxidation

○長尾 遼, 中西 華代, 野口 巧（名古屋大院・理）

Ryo Nagao, Hanayo Ueoka-Nakanishi, Takumi Noguchi (*Grad. Sch. Sci., Univ. Nagoya*)

15:15～16:15　O 会場（203 教室）／Room O (203)
2Ob　核酸：構造・物性 / Nucleic acid

2O1515　**DNA の結晶構造中の構造ゆらぎの解析**

Reconsidered DNA conformations in crystal structures

○角南 智子, 河野 秀俊（原子力機構）

Tomoko Sunami, Hidetoshi Kono (*JAEA*)

2O1530　**Temperature Dependence of Structural Dynamics of RNA and DNA Hairpins Studied by 2D Fluorescence Lifetime Correlation Spectroscopy**

Chao-Han Cheng[1], Kunihiko Ishii[1,2], Tahei Tahara[1,2] ([1]*Molecular Spectroscopy Laboratory, RIKEN*, [2]*Ultrafast Spectroscopy Research Team, RIKEN Center for Advanced Photonics*)

2O1545　**DNA 酵素による紫外線損傷 DNA 光修復の構造解析**

Structural analysis of DNAzyme that functions as photorepair of UV-damaged DNA

○倉橋 雄飛, 岩田 達也, 神取 秀樹（名古屋工業大学）

Yuhi Kurahashi, Tatsuya Iwata, Hideki Kandori (*Nagoya Inst. Tech*)

2O1600　クロマチンは塩によってどのようにコンパクトになるのか？

How can chromatin condense with salt? - a model study using a synthetic nucleosome system

○前島 一博[1], 引間 孝明[2], Rogge Ryan[3], Hansen Jeffrey[3], 田村 佐知子[1]（[1]国立遺伝学研究所 構造遺伝学研究センター, [2]理研播磨研究所, [3]コロラド大学）

Kazuhiro Maeshima[1], Takaaki Hikima[2], Ryan Rogge[3], Jeffrey Hansen[3], Sachiko Tamura[1] ([1]*National Institute of Genetics*, [2]*RIKEN SPring-8 Center*, [3]*Colorado State University*)

13:55～16:15　P 会場（205 教室）／Room P (205)
2P　数理生物学 / Mathematical biology

2P1355　多様な昆虫の翅外形を記述する力学モデル

A mechanical model for diversified wing margin shapes among insects

○石本 志高（秋田県立大学 システム科学技術学部）

Yukitaka Ishimoto (*Fac. of Mach. Int. & Sys. Eng., Akita Pref. U.*)

2P1410　数理モデルとイメージングを用いた多階層連結による走電性の理解

Integration of Multilayer Stages in ElectroTaxis By Using Live Imageing and Simulation

○安井 真人[1], 松岡 里実[1], 上田 昌宏[1,2]（[1]理化学研究所, [2]大阪大学）

Masato Yasui[1], Satomi Matsuoka[1], Masahiro Ueda[1,2] ([1]*RIKEN*, [2]*Osaka University*)

2P1425　増殖集団における細胞状態の重要性を評価する 1 細胞統計手法

Single-cell statistics to evaluate the significance of cellular state in growing population

○野添 嵩[1], 若本 祐一[1,2]（[1]東大院・総合文化, [2]複雑系生命システム研究センター）

Takashi Nozoe[1], Yuichi Wakamoto[1,2] ([1]*Grad. Sch. Arts and Sci., Univ. Tokyo*, [2]*Research Center for Complex Systems Biology, Univ. Tokyo*)

2P1440　簡単な細胞モデルを用いた Pirt 方程式の導出

Derivation of the Pirt equation in the simple cellular model

○姫岡 優介, 金子 邦彦（東大総合文化）

Yusuke Himeoka, Kunihiko Kaneko (*Tokyo Univ. Department of Arts and Sciences*)

2P1455　遺伝子発現及びエピジェネティック修飾ダイナミクス間のタイムスケール

The time scale between gene expression and epigenetic modification dynamics

○宮本 直, 金子 邦彦（東大・総文）

Tadashi Miyamoto, Kunihiko Kaneko (*Grad. Sch. Art. Sci., Univ. Tokyo*)

休憩 15:10-15:15

2P1515　ネットワーク構造のデザイン原理と構成要素の応答性

Network Design Principles and Response Sensitivity of Components

○井上 雅世[1], 金子 邦彦[2]（[1]産総研 molprof, [2]東大 総合文化）

Masayo Inoue[1], Kunihiko Kaneko[2] ([1]*molprof, AIST*, [2]*Univ. of Tokyo*)

2P1530　触媒反応ネットワークにおける資源不足と多様化

Diversification by limitation of multiple resources in a catalytic reaction network

○上村 淳, 金子 邦彦（東京大学大学院総合文化研究科）

Atsushi Kamimura, Kunihiko Kaneko (*Dept. of Basic Science, The University of Tokyo*)

2P1545　全成分タンパク質合成反応モデルの構築とこれを用いた反応ダイナミクス解析

Reaction dynamics analysis of the whole protein translation system by computational modeling

○松浦 友亮[1], 清水 義宏[2], 細田 一史[3], 谷村 直樹[4], 四方 哲也[5]（[1]大阪大学 工学研究科, [2]理研QBic, [3]大阪大学 未来機構, [4]ミズホ情報総研, [5]大阪大学 情報科学研究科）

Tomoaki Matsuura[1], Yoshihiro Shimizu[2], Kazufumi Hosoda[3], Naoki Tanimura[4], Tetsuya Yomo[5] ([1]*Department of Biotechnology, Osaka University*, [2]*Qbic, RIKEN*, [3]*Institute for Academic Initiatives, Osaka University*, [4]*Mizuho Information and Research Institute*, [5]*Department of Bioinformatic Engineering*)

2P1600　Polynomial-life model towards analysis of turnover and regeneration

Hiroshi Yoshida (*Kyushu Univ. Dept. Math.*)

13:55～15:10　Q 会場（207 教室）／Room Q (207)
2Q　生体膜・人工膜 II / Biological & Artificial membrane II

2Q1355　植物の傷害応答性・長距離・高速カルシウムシグナル伝達

Mechanical wounding/insect attack-induced, long-distance, rapid calcium signal transduction in plants

○豊田 正嗣[1,2], Gilroy Simon[1]（[1]University of Wisconsin-Madison, [2]JST・さきがけ）

Masatsugu Toyota[1,2], Simon Gilroy[1] ([1]*University of Wisconsin-Madison*, [2]*JST, PRESTO*)

2Q1410　パターン化モデル生体膜を用いた、光シグナル伝達に関わる膜タンパク質の脂質ラフト親和性解析

Raftophilicity of membrane proteins in the phototransduction cascade evaluated in a patterned model membrane

○谷本 泰士[1], 小嶋 佐妃子[1], 森垣 憲一[1,2], 林 文夫[3]（[1]神戸大学農学研究科生命機能科学専攻, [2]神戸大自然科学先端融合環遺伝子実験センター, [3]神戸大学理学研究科）

Yasushi Tanimoto[1], Sakiko Kojima[1], Kenichi Morigaki[1,2], Fumio Hayashi[3] ([1]*Graduate School of Agricultural Science, Kobe University*, [2]*Organization of Advanced Science and Technology Research Center for Environmental Genomics, Kobe Univ.*, [3]*Graduate School of Science, Kobe University*)

2Q1425　Molecular Gate Locations in MthK Potassium Channels

Crina Nimigean (*WCMC*)

2Q1440　全反射赤外分光で見る電位依存性プロトンチャネル VSOP への金属結合

Metal binding to the voltage-gated proton channel VSOP studied by ATR-FTIR

○岩城 雅代[1,2], 竹下 浩平[3,4,5], 岡村 康司[2,6], 中川 敦史[2,3], 神取 秀樹[1,2]（[1]名工大, [2]JST-CREST, [3]阪大 蛋白研, [4]阪大 未来戦略機構, [5]JST-さきがけ, [6]阪大院 医）

Masayo Iwaki[1,2], Kohei Takeshita[3,4,5], Yasushi Okamura[2,6], Atsushi Nakagawa[2,3], Hideki Kandori[1,2] ([1]*Nagoya Inst. Tech.*, [2]*JST-CREST*, [3]*Inst. Protein Res., Osaka Univ.*, [4]*Inst. Acad. Initiat., Osaka Univ.*, [5]*JST-PRESTO*, [6]*Grad. Sch. Med., Osaka Univ.*)

2Q1455　ナトリウムポンプロドプシンの電気生理学

Electrophysiology of Na^+ pumping rhodopsins

○角田 聡[1,2], 神取 秀樹[1,2]（[1]名工大 院工, [2]名工大 オプトバイオテクノロジー研究センター）

Satoshi Tsunoda[1,2], Hideki Kandori[1,2] ([1]*Nagoya Institute of Technology*, [2]*OptoBioTechnology Research Center*)

ポスター　Poster

第 1 日目（9 月 13 日（日））／Day 1（Sep. 13 Sun.）プロムナード / Promenade

蛋白質：構造 / Protein: Structure

1Pos001 **電子顕微鏡を用いた核ラミンの動的構造変化**
Observation of dynamics of nuclear lamin using electron microscopy
Muneyo Mio[1], Mai Tsunoda[1], Hayato Yamashita[2], Toshihiko Sugiki[3], **Kazuhiro Mio**[1] ([1]*AIST*, [2]*Grad. Sch. Eng. Sci., Univ. Oosaka*, [3]*IPR, Univ. Oosaka*)

1Pos002 **Single-particle analysis of *Thermus thermophilus* V-ATPase using an electron microscopy**
Atsuko Nakanishi[1], Nao Takeuchi[1], Jun-ichi Kishikawa[1], Kaoru Mitsuoka[2], Ken Yokoyama[1] ([1]*Kyoto Sangyo Univ. LifeSci.*, [2]*Osaka Univ. Res. Ctr. UHVEM*)

1Pos003 **分子動力学法と分子ドッキング法を用いたエストロゲンレセプターのリガンド結合予測**
Prediction of binding pose of estradiol to human estrogen receptor: identification of druggable pocket and ensemble-based docking
Hiroaki Saito, Hidemi Nagao, Kazutomo Kawaguchi (*Kanazawa University*)

1Pos004 **クライオ電子顕微鏡を用いた B 型肝炎ウイルスの表面抗原粒子の構造解析**
Structural analysis of hepatitis B surface antigen particles with cryo-electron microscopy
Mai Tsunoda[1], Muneyo Mio[1], Yasuko Maeda[2], Yuji Hoshi[3], Kaoru Mitsuoka[4], Kazuhiro Mio[1] ([1]*AIST*, [2]*arroba.LLC*, [3]*Central Blood Inst., JRCS*, [4]*Research Center for UHV-EM, OSAKA Univ.*)

1Pos005 **NLRP3 タンパク質リガンド結合ドメインの構造特性**
Structure characterization of the ligand-binding domain of an inflammation-related protein NLRP3
Ryota Yamamoto[1], Kazuto Yamashita[1], Hiroshi Imamura[2], Motonari Tsubaki[1], Eri Chatani[1] ([1]*Grad. Sch. Sci.,Kobe Univ.*, [2]*AIST*)

1Pos006 **コレラ菌走化性受容体のリガンド認識機構**
Ligand recognition mechanism of chemoreceptor proteins of *Vibrio cholerae*
Yohei Takahashi[1], Kazumasa Sumita[1], Yumiko Uchida[1], So-ichiro Nishiyama[2], Ikuro Kawagishi[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci. Osaka Univ.*, [2]*Dept. Front. Biosci. Sci., Hosei Univ.*)

1Pos007 **EMDB と PDB データの形状類似検索: Omokage 検索**
Shape similarity search for EMDB and PDB: Omokage search
Hirofumi Suzuki[1,2], Takeshi Kawabata[1], Haruki Nakamura[1,2] ([1]*IPR, Osaka-univ*, [2]*PDBj*)

1Pos008 **Porphyromonas gingivalis の T9SS によって分泌される PGN_0123 の結晶化**
Crystallization of PGN_0123 secreted by T9SS of Porphyromonas gingivalis
Yusuke Handa[1], Keiko Sato[2], Koji Nakayama[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci. Osaka Univ*, [2]*Grad. Sch. Biomedical Sci., Univ. Nagasaki*)

1Pos009 **レジストリ―の異なるダイニンストーク領域の二次構造解析**
Secondary structure analysis of the entire stalk region with two different registries
Haruka Iwasaki[1], Yosuke Nishikawa[2], Momoko Inatomi[1], Hideaki Tanaka[2], Genji Kurisu[2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Institute for Protein Research*)

1Pos010 **水溶性セレノキシド試薬を用いた α-lactalbumin の酸化的フォールディング**
The oxidative folding of α-lactalbumin under mild basic conditions by using a selenoxide reagent
Reina Shinozaki, Michio Iwaoka (*Tokai University*)

1Pos011 **分子動力学シミュレーションを用いた Hras-GTP/GDP 複合体周辺の水の動きの解析**
Analysis of dynamics of water molecules near the Hras-GTP/GDP complexes by molecular dynamics simulations
Takeshi Miyakawa[1], Ryota Morikawa[1], Masako Takasu[1], Kimikazu Sugimori[2], Kazutomo Kawaguchi[2], Hiroaki Saito[2], Hidemi Nagao[2] ([1]*Tokyo Univ. of Pharm. and Life Sci.*, [2]*Kanazawa Univ.*)

1Pos012 **プロテイン A とその誘導体の溶液内構造。X 線小角産卵法による研究。**
Structure of Protein A and its derivative in solution studied by small-angle x-ray scattering
Masaji Shinjo[5], Kaoru Ishimura[6], Akitsugu Yamamoto[6], Hiroshi Kihara[1,2,3,4] ([1]*Himeji Hinomoto College*, [2]*Hinomoto Gakuen Educational Foundation*, [3]*Ritsumeikan Univ. SR Center*, [4]*Nagoya Univ. SR Research Center*, [5]*Kansai Medical Univ*, [6]*Nagahama Inst of Bio-Sci. & Tech*)

1Pos013 **分子動力学シミュレーションによる Aβ アミロイド線維の揺らぎと構造**
Structure and fluctuation of Aβ fibril by molecular dynamics simulations
Hisashi Okumura[1,2], Satoru G. Itoh[1,2] ([1]*IMS*, [2]*SOKENDAI*)

1Pos014 **tRNA チオ化酵素 TtuA-TtuB 複合体の結晶構造解析**
Crystal structure analysis of TtuA-TtuB, a tRNA thiolation enzyme complex
Shun Narai[1], Minghao Chen[1], Naoki Shigi[3], Yoshikazu Tanaka[2], Min Yao[2] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Facl. of Adv. Life Sci., Hokkaido Univ.*, [3]*Biotech. Res. Inst. for Drug Discov., AIST*)

1Pos015 **The approach toward crystallization of HBV core of genotype C**
Katsumi Omagari, Yasuhito Tanaka (*Dept. Virol., Grad Sch. Med., Nagoya City Univ.*)

1Pos016 **Development of a De Novo protein structure prediction: generating new fold structures by permutating and reversing SSEs of known folds**
Shunsuke Nishiyama, Kodai Takagi, George Chikenji (*Grad. Sch. of Engineering, Nagoya Univ*)

1Pos017 **コフィリンの結合によるアクチン線維構造変化の解析**
Elucidation of structural change in actin filament invoked by cofilin
Kotaro Tanaka[1], Chieko Kimura-Sakiyama[1], Kaoru Mitsuoka[2], Daisuke Kasuya[3], Yuichiro Maeda[1], Akihiro Narita[1] ([1]*Structure Biology Research Center, Nagoya Univ.*, [2]*Research Center for Ultra-High Voltage Electron Microscopy, Osaka Univ.*, [3]*JBiC*)

蛋白質：構造･機能 / Protein: Structure & Function

1Pos018 **MP2/6-311G++(d,p)法による炭酸脱水酵素の機能発現と構造に関する理論的研究**
Basis Set Having Diffuse Function, 6-311G++(d,p), in MP2 to Calculate Energy of Active Site Model Containing His/Trp in Carbonic Anhydrase
Muhamad Koyimatu[1], Kimikazu Sugimori[2], Hidemi Nagao[2], Hideto Shimahara[1] ([1]*Japan Advanced Institute of Science and Technology*, [2]*Kanazawa University*)

1Pos019 **多剤排出トランスポーター AcrB の Motion-Tree 法による解析**
Motion tree analysis of the multidrug transporter AcrB
Tsutomu Yamane[1], Ryotaro Koike[2], Motonori Oota[2], Satoshi Murakami[3], Akinori Kidera[1], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [2]*Grad. Sch. of Inf. Sci., Nagoya Univ.*, [3]*Grad. Sch. of Biosci. and Bioeng., Tokyo Inst. of Tech.*)

1Pos020 **シアノバクテリア由来のフィトクロム Cph1 の光受容部位の構造変化ダイナミクス**
Conformational dynamics of photosensory domains of the Cyanobacterial Phytochrome 1 (Cph1)
Kimitoshi Takeda, Masahide Terazima (*Grad. Sch. Sci., Kyoto Univ*)

1Pos021 **アメリカヤマゴボウ抗ウイルスタンパク質におよぼす糖結合の影響**
Sugar binding effects on pokeweed anti-viral protein
Ayana Okuno[1], Ryosuke Ueno[2], Yuki Okada[2], Etsuko Nishimoto[3] ([1]*Sch. Agr.,Univ. Kyushu*, [2]*Grad. Sch. Bioresour. Bioenveron. Sci.,Univ. Kyushu*, [3]*Fac. Agr.,Univ.Kyushu*)

1Pos022 **密度汎関数法による D-アミノ酸酸化酵素のフラビン-基質配置に関する研究**
A DFT study on flavin-substrate arrangement in D-amino acid oxidase
Kyosuke Sato (*Dept. Mol. Physiol., Facult. Life Sci., Kumamoto Univ.*)

1Pos023 **26s プロテアソームの高速 AFM 観察**
High-speed AFM observation of the 26S proteasome
Takashi Okuno[1], Kentaro Noi[2,5], Ken-ichi Arita-Morioka[2,5], Hikaru Tsuchiya[3], Yasushi Saeki[3], Kazunobu Takahashi[4], Tomonao Inobe[4], Takahiro Saito[1], Kunitoshi Yamanaka[2,5], Teru Ogura[2,5] ([1]*Fac. Sci. Univ. Yamagata.*, [2]*Inst. Mol. Embryol. Genet., Kumamoto Univ.*, [3]*Tokyo Metro. Inst. of Med. Sci.*, [4]*Frontier Res. Core for Life Sci., Univ. of Toyama*, [5]*CREST, JST*)

1Pos024 **Structural origins of slowness and regulatory mechanism in KaiC ATPase**
Jun Abe[1], Takuya B. Hiyama[1], Atsushi Mukaiyama[1,2], Seyoung Son[3], Toshifumi Mori[2,4], Shinji Saito[2,4], Masato Osako[3], Wolanin Julie[1], Eiki Yamashita[5], Takao Kondo[3], Shuji Akiyama[1,2] ([1]*CIMoS, IMS*, [2]*Grad. Univ. for Advanced Studies*, [3]*Div. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*, [4]*Dept. Theoret. and Comput. Molecular Science, IMS*, [5]*IPR*)

1Pos025 **Study of interaction between transferrin and transferrin receptor2 by molecular simulation and flow cytometry**
Tetsuya Sakajiri[1], Katsuya Ikuta[2], Takaki Yamamura[1] ([1]*Morioka Univ.*, [2]*Asahikawa Med. Col.*)

1Pos026 **植物型フェレドキシンの酸化還元電位を大幅に上昇させる構造要因の精密解析**
Structural basis for the unexpected increase of redox potential of plant-type Ferredoxin revealed by the high resolution X-ray analysis
Daiki Kiyota[1,2], Arisa Sato[3], Risa Mutoh[2], Haruki Yamamoto[2], Toshiharu Hase[1,2], Genji Kurisu[1,2] ([1]*Grad. Sch. Sci. Osaka Univ.*, [2]*Institute for Protein Research, Osaka University, Japan*, [3]*Doshisha Women's college*)

1Pos027 **蛋白質多重配列アラインメントの格子気体模型**
Lattice gas model of protein multiple sequence alignment
Akira Kinjo (*Inst. Protein Res., Osaka Univ.*)

1Pos028 **Analysis of the Free Energy Landscapes for the Open-Closing Dynamics of MalK2 Using Enhanced Sampling MD Simulation**
WeiLin Hsu, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech.*)

1Pos029 **How do intramolecular hydrogen bonds within a peptide contribute to peptide-antibody interaction?**
Kazuhiro Miyanabe[1], Hiroki Akiba[2], Jose Caaveiro[2], Daisuke Kuroda[2], Makoto Nakakido[3], Osamu Arai[4], Hiroko Iwanari[4], Takao Hamakubo[4], Kouhei Tsumoto[1,2,3] ([1]*Dept. Chem. Biotech., Sch. Eng., Univ. Tokyo*, [2]*Dept. Bioeng., Sch. Eng., Univ. Tokyo*, [3]*Inst. Med. Sci., Univ. Tokyo*, [4]*RCAST, Univ. Tokyo*)

1Pos030 **Molecular dynamics of channelrhodopsin at the early stages of channel opening**
Mizuki Takemoto[1], E. Hideaki Kato[1], Michio Koyama[1], Jumpei Ito[2], Motoshi Kamiya[3], Shigehiko Hayashi[3], Andres D. Maturana[2], Karl Deisseroth[4], Ryuichiro Ishitani[1], Osamu Nureki[1] ([1]*University of Tokyo*, [2]*Nagoya University*, [3]*Kyoto University*, [4]*Stanford University*)

1Pos031 **滴定 X 線小角散乱測定による Shootin1-Cortactin 複合体形成の構造学的評価**
Structural investigation of direct interaction between Shootin1 and cortactin by the titration SAXS measurements
Junko Makino[1], Hironari Kamikubo[1], Yoichi Yamazaki[1], Mikio Kataoka[1], Keito Yoshida[1], Naoyuki Inagaki[2], Yusuke Kubo[2], Kentarou Baba[2] ([1]*Graduate School of Materials Science, Nara Institute of Science and Technology*, [2]*Graduate School of Biological Sciences, Nara Institute of Science and Technology*)

1Pos032 **Proton transfer mechanisms of photosystem II: Hybrid ab inito quantum mechanics study**

Atsushi Nakamura[1], Jiyoung Kang[1], Yasufumi Umena[2], Keisuke Kawakami[3], Shen Jian-Ren[2], Nobuo Kamiya[3], Masaru Tateno[1] ([1]*Grad. Sch. Life Sci.,Univ. Hyogo*, [2]*Grad. Sch. of Natural Science and Technology, Okayama Univ*, [3]*OCARINA, Osaka City Univ.*)

1Pos033 **分子動力学シミュレーションを用いた細菌機械受容チャネル MscL の脂質膜厚変化で影響される開口過程の解析**

Molecular Dynamics Study on the Gating of the Bacterial Mechanosensitive Channel MscL Affected by Membrane Thickness

Hiroki Katsuta[1], Yasuyuki Sawada[2], Masahiro Sokabe[3] ([1]*Scl. Med. Nagoya Univ.*, [2]*Dept. Physiol. Grad. Scl. Med. Nagoya Univ.*, [3]*Mechanobiology Lab. Grad. Scl. Med. Nagoya Univ.*)

1Pos034 **Structure and dynamics of Sec protein-conducting channel**

Yasunori Sugano[1], Yoshiki Tanaka[1], Mizuki Takemoto[2], Takaharu Mori[3], Takamitsu Haruyama[4], Arata Furukawa[1], Tsukasa Kusakizako[2], Kaoru Kumazaki[2], Ayako Kashima[2], Ryuichiro Ishitani[2], Hiroki Konno[4], Yuji Sugita[3], Osamu Nureki[2], Tomoya Tsukazaki[1,5] ([1]*NAIST*, [2]*Grad. Sch. of Sci., Univ. of Tokyo*, [3]*RIKEN*, [4]*BioAFM-FRC. Kanazawa Univ*, [5]*JST, PRESTO*)

1Pos035 **含セレン酵素の活性中心を模倣した短鎖セレノペプチドの分子設計**

Molecular design of short selenopeptides mimicking a selenoenzyme active center

Natsuki Babe, Toshiki Suzuki, Taku Shimosato, Toshiya Minezaki, Michio Iwaoka (*School Sci., Tokai Univ.*)

蛋白質：特性 / Protein: Property

1Pos036 **内部座標系による構造エントロピー変化の計算と解析**

Analysis for configurational entropy change calculated using internal coordinate system

Simon Hikiri[1], Takashi Yoshidome[2], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. of Med. Life Sci. Yokohama city Univ.*, [2]*Dept. of Appl. Phys., Tohoku Univ.*)

1Pos037 **超音波連続照射下におけるマウスプリオン蛋白質の凝集体形成**

The formation of aggregates of mouse prion protein under the continuous ultrasonic irradiation

Kei-ichi Yamaguchi[1], Ryo P. Honda[1,2], Abdelazim Elsayed Elhelaly[1], Kazuo Kuwata[1,2] ([1]*Unit. Grad. Sch. of Drug Dis. and Med. Inf. Sci., Gifu Univ.*, [2]*Grad. Sch. of Med., Gifu Univ.*)

1Pos038 **SyPixD の光反応に対する構造揺らぎの効果**

Effect of structural fluctuation on photoreaction of SyPixD

Tsubasa Nakajima[1], Kunisato Kuroi[1], Kouji Okajima[2,3], Masahiko Ikeuchi[3], Satoru Tokutomi[2], Masahide Terazima[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Osaka Pref Univ.*, [3]*Grad. Sch. Art and Sci., Tokyo Univ.*)

1Pos039 **アルコール脱水素酵素のサブユニット間相互作用に及ぼすホフマイスター効果**

Hofmeister effect on the subunit-subunit interaction of Liver Alcohol Dehydrogenase

Tomohiro Aoyama[1], Etsuko Nishimoto[2] ([1]*Grad. Sch. Bioresour. Bioenviron. Sci., Univ. Kyushu*, [2]*Fac. Agr., Univ. Kyushu*)

1Pos040 **ユビキチン化の新規物理化学的性質**

Novel physicochemical properties of ubiquitylation

Daichi Morimoto[1], Erik Walinda[1], Kenji Sugase[1], Harumi Fukada[2], Yu-shin Sou[3], Shun Kageyama[3,4], Masaru Hoshino[5], Takashi Fujii[6], Hikaru Tsuchiya[7], Yasushi Saeki[7], Kyohei Arita[8], Mariko Ariyoshi[1], Hidehito Tochio[9], Kazuhiro Iwai[10], Keiichi Namba[6,11], Masaaki Komatsu[3,4], Keiji Tanaka[7], Masahiro Shirakawa[1] ([1]*Eng., Kyoto Univ.*, [2]*Life Envi. Sci., Osaka Pref. Univ.*, [3]*Protein Metabolism Proj., Tokyo Metro. Ins. Med. Sci.*, [4]*Med., Niigata Univ.*, [5]*Pharm., Kyoto Univ.*, [6]*QBiC, RIKEN*, [7]*Lab. Protein Metabolism, Tokyo Metro. Ins. Med. Sci.*, [8]*Med. Life Sci., Yokohama City Univ.*, [9]*Science, Kyoto Univ.*, [10]*Med., Kyoto Univ.*, [11]*Frontier Biosci., Osaka Univ.*)

1Pos041 **時間分解 EPR 法を用いたアミロイド線維形成過程におけるヒトインスリン局所構造変化の観測**

Time-resolved EPR Study on Local Structures of Human Insulin Forming Amyloid Fibrills

Tomoka Abe, Takashi Tachikawa, Eri Chatani, Yasuhiro Kobori (*Grad. Sch. Sci., Kobe Univ.*)

1Pos042 **膜貫通 β バレルの β ストランドのねじれと曲りは抑制されている**

β-strand twisting and bending of the transmembrane β-barrel are suppressed

Nobuaki Kikuchi, Shinichi Ebisawa, Yuka Watanabe, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. of Bioinformatics, Soka Univ.*)

1Pos043 **複数のアミロイド性ペプチドの混在する複雑な系におけるアミロイド線維形成機構**

Amyloid Fibrillation in Complicated Systems Containing Various Amyloidogenic Peptides

Hiroya Muta[1], Masatomo So[1], Kazumasa Sakurai[2], Yuji Goto[1] ([1]*Inst. for Pro. Research, Osaka Univ.*, [2]*Inst. of Adv. Tech., Kinki Univ.*)

1Pos044 **Study on the NaCl concentration-dependent structural stability of maize (leaf) ferredoxin**

Misaki Kinoshita, Yoko Ariga, Toshiharu Hase, Yuji Goto, Genji Kurisu, Young-Ho Lee (*Institute for Protein Research, Osaka University*)

1Pos045 **抗がん剤候補のハースタチンは天然変性タンパク質である**

Herstatin, an antitumor drug candidate, is an intrinsically disordered protein

Daisuke Tashiro, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

1Pos046 **GB1(41-56)ペプチドの β ヘアピン構造に及ぼす圧力効果に関する FTIR・ラマン分光研究**

FTIR and Raman studies of pressure effects on the β-hairpin structure of GB1(41-56) peptides

Minoru Kato[1], Keita Tsuchiya[2] ([1]*Dept. Pharmacy. Ritsumeikan Univ.*, [2]*Grad. Sch. Life Sci. Ritsumeikan Univ.*)

蛋白質：機能 / Protein: Function

1Pos047 **Control of protein aggregation and oligomerization using short SEP (Solubility Enhancing Peptide) tags**

Md. Golam Kabir, Mohammad Monirul Islam, Yutaka Kuroda (*Tokyo Univ. Agri. Eng.*)

1Pos048 ダイナミン 1-コルタクチンおよびダイナミン 1–アンフィファイジン複合体の高速 AFM による動態観察
High-Speed AFM imaging of dynamics of Dynamin 1-Cortactin and Dynamin 1-Amphiphysin 1 complexes
Yusuke Kumagai[1], Tetsuya Takeda[3], Takayuki Uchihashi[1,2], Tadashi Abe[3], Kohji Takei[3], Toshio Ando[1,2] ([1]*College of Science and Engineering, Kanazawa University*, [2]*Bio-AFM Frontier Research Center, College of Science and Engineering, Kanazawa University*, [3]*Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama University*)

1Pos049 線虫ミトコンドリアにおけるフェレドキシン依存性電子伝達経路の機能解析
Characterization of ferredoxin-dependent electron transfer system in Caenorhabditis elegans mitochondira
Ryohei Wada[1], Hiroshi Hori[1], Yoshito Furuie[1], Fusako Takeuchi[2], Motonari Tsubaki[1] ([1]*Grad. Sch. Sci., Chem., Kobe Univ.*, [2]*IPHE., Kobe Univ.*)

1Pos050 ラン藻でのアルカン合成に必要な 2 つの酵素間の結合部位の探索
The search for the binding sites between two enzymes essential for cyanobacterial alkane biosynthesis
Mari Chang[1], Yuuki Hayashi[2], Munehito Arai[1,2] ([1]*Dept. Phys., Univ. Tokyo*, [2]*Dept. Life Sci., Univ. Tokyo*)

1Pos051 アルカン合成酵素のアラニンスキャン変異解析
Residues essential for the alkane producing activity of aldehyde deformylating oxygenase revealed by alanine scanning mutagenesis
Keigo Shimba, Fumitaka Yasugi, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

1Pos052 海洋性ビブリオ菌のべん毛本数を負に制御する FlhG の蛋白質凝集性と ATPase 活性の相関
Relationship between aggregability and ATPase activity of FlhG, the negative regulator of the flagellar number in *Vibrio alginolyticus*
Hikaru Hirata, Michio Homma, Seiji Kojima (*Div. Bio Sci., Grad. Sch. Sci., Univ. Nagoya*)

蛋白質：計測･解析 / Protein: Measurement & Analysis

1Pos053 **4. Development of a new BRET based ATP indicator for high-throughput quantitative ATP assay**
Tomoki Yoshida, Hiromi Imamura (*Graduate school of biostudy, Kyoto University*)

1Pos054 難溶性化合物とタンパク質との相互作用解析へ向けた新規技術開発
A novel approach for analysis of interaction between low water-soluble compounds and target proteins
Shigeru Sugiyama[1,2], Keisuke Kakinouchi[4], Hiroyoshi Matsumura[3,4], Hiroaki Adachi[3,4], Kazufumi Takano[4,5], Mihoko Maruyama[3], Yoshinori Takahashi[3], Hiroshi Yoshikawa[6], Masashi Yoshimura[3], Satoshi Murakami[4,7], Tsuyoshi Inoue[3,4], Michio Murata[1,2], Yusuke Mori[3,4] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*JST ERATO*, [3]*Grad. Sch. Eng., Osaka Univ.*, [4]*SOSHO Inc.*, [5]*Grad. Sch. Life Environ. Sci., Kyoto Pref. Univ.*, [6]*Grad. Sch. Sci. & Eng., Saitama Univ.*, [7]*Grad. Sch. Biosci. & Biotech., Tokyo Inst. Tech.*)

1Pos055 X 線 1 分子追跡法よるフェムトニュートン力場でのタンパク質ネットワーク観察
Observation of Supersaturated Protein Networking in femto-Newton Force-field from Diffracted X-ray Tracking
Yufuku Matsushita[1], Hiroshi Sekiguchi[2], Noboru Ohta[2], Keigo Ikezaki[1], Yuji Goto[3], Yuji C. Sasaki[1,2] ([1]*Graduate School of Frontier Science, The University of Tokyo*, [2]*SPring-8/JASRI*, [3]*Institute for Protein Research, Osaka University*)

1Pos056 アカネ科植物由来 抗腫瘍活性ペプチド RA-VII の分子動力学シミュレーション
Molecular dynamics simulations of antitumor peptide RA-VII from Rubia cordifolia
Yoh Noguchi[1], Hironao Yamada[1], Sakiko Mori[1], Takeshi Miyakawa[1], Ryota Morikawa[1], Satoshi Yokojima[2], Yukio Hitotsuyanagi[2], Koichi Takeya[2], Masako Takasu[1] ([1]*School of Life Sciences, Tokyo University of Pharmacy and Life Sciences*, [2]*School of Pharmacy, Tokyo University of Pharmacy and Life Sciences*)

1Pos057 解析ソフトウェア CAPAXIS のさらなる開発：PyMOL との連携と B 型肝炎ウイルスへの応用
Further Development of Software Tool, CAPAXIS, for Use with PyMOL : Application to Hepatitis B Virus
Shunsuke Sato[1], Go Watanabe[2], Shigetaka Yoneda[2] ([1]*Grad. Sch. Sci., Kitasato Univ.*, [2]*Sch. Sci., Kitasato Univ.*)

蛋白質：工学 / Protein: Engineering

1Pos058 新型マイクロアレイ MMV を用い機能性ペプチドを淘汰するのに初めて成功したーセルベーストスクリーニングの躍進
First success in selection of functional peptides using a novel concept microarray MMVーA breakthrough for cell-based screenings
Koichi Nishigaki[1,2], Tatsuya Furukawa[1], Naoki Takeuchi[1], Takuto Saiki[1], Aya Hongo[1], Motoki Iwano[1], Yuuki Masubuchi[1], Miho Suzuki[1] ([1]*Saitama Univ.*, [2]*AIST*)

1Pos059 主鎖二面角パターンによる αβ 型タンパク質構造のデザイン
Design of αβ protein structures: the extended design principles
Yu-Ru Lin[1], Rie Koga[1,3,4], Gaohua Liu[2], Guy Montelione[2], David Baker[1], **Nobuyasu Koga**[1,3,4] ([1]*Univ. Wash.*, [2]*Rutgeres, NESG*, [3]*IMS, CIMoS*, [4]*JST, PRESTO*)

1Pos060 合理的設計による抗体精製用リガンド FPA の抗体解離 pH の向上
Rational design to improve the pH-sensitive antibody dissociation of FPA, a ligand for antibody purification
Taihei Sawada, **Yoshiki Oka**, Takahiro Watanabe, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

1Pos061 Rossmann フォールド蛋白質のデザイン：ストランド入れ替わり問題の謎に迫る
Design of Rossmann folds: the internal strand swapping problem
Rie Koga[1,2], Gaohua Liu[3], Gaetano T. Montelione[3], David Baker[2], Nobuyasu Koga[1,2,4] ([1]*IMS, CIMoS*, [2]*Univ. of Washington, Dept. Biochem.*, [3]*Rutgers Univ., NESG*, [4]*JST, PRESTO*)

ヘム蛋白 / Heme proteins

1Pos062 **電気化学マイクロデバイスによる P450 反応解析**
Electrochemical Analysis of P450s in Microfluidic Channel
Yasuhiro Mie, Yasuo Komatsu (*Bioproduction Res. Inst., AIST*)

1Pos063 **ヘムの構造歪みが酸化還元電位に与える影響の計算科学的研究**
Computational Study of Structural Effects on Redox Potential of Hemes
Yasuhiro Imada[1,2], Haruki Nakamura[1], Yu Takano[1,3] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Grad. Sch. Info. Sci., Hiroshima City Univ.*)

1Pos064 **Bach2 天然変性領域の電荷状態分布の解析**
Charge-state-distribution analysis of Bach2 intrinsically disordered heme binding region
Tomoji Suenaga[1], Miki Watanabe-Matsui[2,3], Hiroki Shima[3], Kazuhiko Igarashi[3], Kazutaka Murayama[4] ([1]*ThermoFisherScientific*, [2]*Tohoku Univ., NICHE*, [3]*Tohoku Univ., Med.*, [4]*Tohoku Univ., Biomed. Eng.*)

1Pos065 **チトクロム酸化酵素を用いた常温高分解能 X 線回折実験法の確立**
Development of high-resolution X-ray diffraction experiments at room temperature
Keita Hatano[1], Akari Miyamoto[1], Atsuhiro Shimada[1], Seiki Baba[2], Takashi Kumasaka[2], Kyoko Shinzawa-Itoh[1], Tomitake Tsukihara[1,3], Shinya Yoshikawa[1] ([1]*Picobiol. Inst., Grad. Sch. Life Sci., Univ. Hyogo*, [2]*JASRI*, [3]*Inst. Protein Res., Osaka Univ.*)

1Pos066 **Electrochemical behavior of bacterial nitric oxide reductase immobilized on gold electrodes *via* self-assembled monolayers**
Kuniaki Yamaki[1], Masaru Kato[1], Takehiko Tosya[2], Ichizo Yagi[1] ([1]*Grad. Sch. Env. Sci., Univ. Hokkaido*, [2]*Harima Inst., Riken*)

膜蛋白質 / Membrane proteins

1Pos067 **固体 NMR によるフォボロドプシン-トランスデューサー複合体の細胞質側での相互作用の解析**
Analysis of interactions of phoborhodopsin-transducer complex in cytoplasmic side by solid-state NMR spectroscopy
Satoshi Nakatani[1], Yoshiteru Makino[1], Ryota Nishikawa[1], Izuru Kawamura[1], Naoki Kamo[2], Akira Naito[1] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Hokkaido Univ.*)

1Pos068 **GPCR の熱安定性を向上させるアミノ酸置換の予測:有力な手法の構築**
Prediction of Thermostabilizing Mutations for G Protein-Coupled Receptors: Construction of an Efficient Method
Yuta Kajiwara[1], Satoshi Yasuda[2], Yuuki Takamuku[3], Nanao Suzuki[3], Takeshi Murata[3], Masahiro Kinoshita[2] ([1]*Graduate School of Energy Science, Kyoto University*, [2]*Institute of Advanced Energy, Kyoto University*, [3]*Graduate School of Science, Chiba University*)

1Pos069 **腸内連鎖球菌 V-ATPase のクライオ位相差電子顕微鏡単粒子構造解析**
Single particle phase-contrast cryo-EM 3D reconstruction of Enterococcus hirae V-ATPase
Kazuyoshi Murata[1], Takeshi Murata[2], Hiroshi Ueno[3], Ryota Iino[4] ([1]*National Institute for Physiological Sciences*, [2]*Dept. Science, Chiba University*, [3]*School of Engineering, The University of Tokyo*, [4]*Okazaki Integrated Bioscience/Institute of Molecular Science*)

1Pos070 **High-resolution cryoEM structural analysis of MotPS stator of the bacterial flagellar motor**
Naoya Terahara, Takayuki Kato, Tohru Minamino, Keiichi Namba (*Grad. Sch. of Frontier Biosciences, Osaka Univ.*)

1Pos071 **Highly sensitive measurement of proton flux mediated by POT**
Shinya Ohdate[1], Tsukasa Kusakizako[2], Shintaro Doki[2], Naoki Soga[1], Ryuichiro Ishitani[2], Rikiya Watanabe[1], Osamu Nureki[2], Hiroyuki Noji[1] ([1]*Department of Applied Chemistry, The University of Tokyo*, [2]*Department of Biological Science, The University of Tokyo*)

1Pos072 **メカノセンシティブチャネルはストレプトマイシンの細胞内への侵入に関与するか？**
Are mechanosensitive channels involved in the entry of streptomycin into the bacterial cell?
Kenichi Hashimoto, Fangzhen Zheng, Kazuhiro Nobata, Sanae Yamazaki, Isamu Yabe, Hisashi Kawasaki (*Dept. Green Sust. Chem., Tokyo Denki Univ.*)

1Pos073 **1 分子イメージングで見る GPCR の多量体化とエンドサイトーシス**
Single-molecule imaging of GPCR oligomerization followed by internalization
Masataka Yanagawa[1], Michio Hiroshima[2], Takahiro Yamashita[3], Yoshinori Shichida[3], Yasushi Sako[1] ([1]*Riken*, [2]*QBiC, Riken*, [3]*Grad. Sch. Sci., Kyoto Univ.*)

1Pos074 **上皮成長因子受容体クラスタリングの超解像定量解析**
A Super-resolved Quantitative Analysis of Epidermal Growth Factor Receptor Clustering
Michio Hiroshima[1,2], Masahiro Ueda[1], Yasushi Sako[2] ([1]*RIKEN QBiC*, [2]*RIKEN*)

核酸結合蛋白質 / Nucleic acid binding proteins

1Pos075 **粗視化 MD-SAXS 法とヌクレオソームへの適用**
Coarse-Grained MD-SAXS method and application to nucleosomes
Yuichi Kokabu[1], Takashi Oda[1], Masaaki Sugiyama[2], Hitoshi Kurumizaka[3], Mamoru Sato[1], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*, [2]*Grad. Sch. Sci., Kyoto Univ.*, [3]*Grad. Sch. Sci. Eng., Waseda Univ.*)

1Pos076 **粗視化シミュレーションによるヘテロクロマチン蛋白質 1 のヌクレオソームへの結合研究**
Heterochromatin protein 1 binding to nucleosomes studied by coarse-grained simulations
Shuhei Watanabe[1], Yuichi Mishima[2], Isao Suetake[2], Shoji Takada[1] ([1]*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ.*, [2]*Inst. Protein Res., Osaka Univ.*)

1Pos077 **高活性 TALE 蛋白質の開発とその応用**
Development and application of improved TALE protein
Kazuho Ikeda, Yoko Terahara, Kenta Sumiyama, Yasushi Okada (*QBiC, RIKEN*)

1Pos078 **粗視化モデルによるダイヌクレオソーム間の相互作用とヒストンテイルの役割**
Dinucleosome structure and role of histone tails by coarse-grained model
Hiroo Kenzaki[1], Shoji Takada[2] ([1]*ACCC, Riken*, [2]*Grad. Sch. Sci., Kyoto Univ.*)

核酸：構造･特性 / Nucleic acid: Structure & Property

1Pos079 **G-quadruplex がさまざまの小分子と相互作用することによる構造変化の反応速度論による研究**
Kinetics and mechanism of conformational changes by interaction of G-quadruplexes and small molecules
Masato Tanigawa, Takafumi Iwaki (*Fac. Med., Oita Univ.*)

1Pos080 **蛍光色素から DNA 修飾した単層カーボンナノチューブへのエネルギートランスファーに関する研究**
Research on energy transfer from fluorescent dyes to DNA-wrapped single-walled carbon nanotubes
Shusuke Oura, Masahiro Ito, Yoshikazu Homma, Kazuo Umemura (*Tokyo Univ. of Sci.*)

1Pos081 **異なる損傷源が引き起こす DNA 二本鎖切断の DMSO による保護作用の比較**
Comparison of the protective effect of DMSO on DNA double-stand break with different injury sources
Masami Noda[1], Yuko Yoshikawa[2], Kenichi Yoshikawa[1], Takahiro Kenmotsu[1], Tadayuki Imanaka[2] ([1]*Doshisha University*, [2]*Ritsmeikan University*)

1Pos082 **超音波パルスによる DNA 切断の促進効果**
Pulsing stimuli of ultrasound causes larger damage on DNA than its CW mode: Single DNA observation on double-strand breaks
Rinko Kubota[1], Yusuke Yamashita[1], Yukihiro Kagawa[1], Yuko Yoshikawa[2], Yoshiaki Watanabe[1], Takahiro Kenmotsu[1], Tadayuki Imanaka[2], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life and Medical Sciences, Univ. Doushisha*, [2]*Laboratory of Environmental Biotechnology Research Organization of Science and Technology, Univ. Ritsumeikan*)

核酸：相互作用･複合体形成 / Nucleic acid: Interaction & Complex formation

1Pos083 **2D DNA nanostructure to arrange positions of functional molecules**
Yuki Matsubara, Akira Suyama (*Univ. of Tokyo, Graduate School of Arts and Sciences*)

1Pos084 **エタノールによって引き起こされる DNA の再帰転移現象**
Reentrant transition on the higher-order structure of DNA with the increase of alcohol concentration
Yuki Oda[1], Yuko Yoshikawa[2], Tadayuki Imanaka[2], Kingo Takiguchi[3], Masato Hayashi[3], Kenichi Yoshikawa[1] ([1]*Graduate School of Life and Medical Science, Doshisha University*, [2]*Department of Biotechnology, College of Life Sciences, Ritsumeikan University*, [3]*Department of Molecular Biology, Graduate School of Science, Nagoya University*)

電子状態 / Electronic state

1Pos085 **Photoactive Yellow Protein に現れる H/D 同位体効果の理論的解析**
Theoretical analysis of H/D isotope effect on Photoactive Yellow Protein
Yusuke Kanematsu[1,2], Yu Takano[1], Masanori Tachikawa[2] ([1]*Grad. Sch. Info., Hiroshima City Univ.*, [2]*Grad. Sch. Nanobio., Yokohama City Univ.*)

水･水和･電解質 / Water & Hydration & Electrolyte

1Pos086 **電解質溶液中におけるマクロアニオンの凝集挙動**
Aggregation behavior of macroanions immersed in electrolyte solution
Takuto Sawayama, Ryo Akiyama (*Kyushu Univ., Dept. Chem.*)

1Pos087 **演題取り消し**

1Pos088 **シミュレーション・データマイニングによるリガンド結合系における水分子の振る舞い**
Simulation-based data-mining approach for water behavior of ligand-binding system
Taku Mizukami[1], Viet Cuong Nguyen[3], Tu Bao Ho[2], Hieu Chi Dam[2] ([1]*Sch. of Materials Sci., JAIST*, [2]*Sch. of Knowledge Sci., JAIST*, [3]*HPC Systems Inc.*)

分子遺伝学･遺伝子発現 / Molecular genetics & Gene expression

1Pos089 **Cell differentiation and Reprogramming: A minimal model**
Ashwin S.S., Masaki Sasai (*Nagoya University, Japan*)

発生･分化 / Development & Differentiation

1Pos090 **多能性幹細胞の分化における１細胞解析**
Single-cell-based analysis of differentiation of pluripotent stem cells
Ayumu Kano[1], Yuta Yamamoto[1], Shogo Nakamura[1], Atsushi Maruyama[1], Olga M De Sousa[2], Masahiro Iwahashi[3], Toshiyuki Akaike[4], Kiyoshi Ohnuma[1] ([1]*Nagaoka University of Technology Bioengineering*, [2]*Faculty of Engineering, Science and Technology National University of East Timor*, [3]*Nagaoka University of Technology Electronics and Information Engineering*, [4]*Tokyo Institute of Technology, Yokohama, Japan 226-8501*)

1Pos091 **植物の中の局所微小管と細胞成長の関係について**
On the Relationship between Local Microtubule and Cell Growth in Plants
Satoru Tsugawa, Chun-Biu Li, Tamiki Komatsuzaki (*Research Institute for Electronic Science*)

筋肉 / Muscle

1Pos092 **Control of Morphological Dynamics of Myoblasts Using Stimulus-Responsive Hydrogels Cross-linked by Host-Guest Interactions**
Marcel Hoerning[1], Masaki Nakahata[2], Akihisa Yamamoto[1], Yoshinori Takashima[2], Akira Harada[2], Motomu Tanaka[1] ([1]*iCeMS, Kyoto University*, [2]*Osaka University*)

1Pos093 **CryoEM structure of muscle thin filament with the tropomyosin and troponin complex**
Yurika Yamada[1], Keiichi Namba[1,2], Takashi Fujii[1,3] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*QBiC, RIKEN*, [3]*JST PRESTO*)

1Pos094 **ナノイメージングによるマウス心臓の *in vivo* サルコメア動態解析**
High-speed live imaging of single sarcomere dynamics in the beating mouse heart *in vivo*
Fuyu Kobirumaki[1], Kotaro Oyama[1,2], Togo Shimozawa[4], Seine Shintani[5], Erisa Hirokawa[1], Takako Terui[3], Shin'ichi Ishiwata[2], Norio Fukuda[1] ([1]*Dept. Cell Physol., Jikei Univ. Sch. Med.*, [2]*Dept. Physics, Waseda Univ.*, [3]*Dept. Anesth., Jikei Univ. Sch. Med.*, [4]*Dept. Life Sci. Med. Biosci., Waseda Univ.*, [5]*Dept. Physics, Univ. Tokyo*)

1Pos095 **F-アクチンの水和状態の円二色性分光と DSC 測定による検討**
Study on the correlation of hydration state, ternary structure and heat capacity of F-actin
Takaya Yamaguchi, Ryotaro Chisima, George Mogami, Makoto Suzuki (*Graduate School of Engineering, Tohoku University*)

1Pos096 **電子顕微鏡によるフォルミン mDia1 のアクチンフィラメント端結合構造の解析**
Structural analysis of binding state of formin/mDia1 to actin filament by electron microscopy
Mizuki Matsuzaki, Akihiro Narita (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos097 **アクチン繊維の骨格形状と滑り運動時に生じる破断の原因**
A cause for breaking of sliding actin filament on myosin
Katsunari Sakamoto, Shigeru Sakurazawa (*Future Univ. Hakodate*)

分子モーター / Molecular motor

1Pos098 **Probing stator-protein dynamics of bacterial flagellar motors**
Chien-Jung Lo, Tsaishun Lin (*Department of Physics, National Central University, Taiwan*)

1Pos099 **Effect of Microtubule Deformation on Kinesin-based Cargo Transportation In Vitro**
Tanjina Afrin[1], Arif Md. Rashedul Kabir[2], Daisuke Inoue[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

1Pos100 **High resolution structural analysis of the flagellar hook of Salmonella**
Peter Horvath[1], Tomoko Miyata[1], Hiroko Takazaki[1], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Riken QBiC*)

1Pos101 **マルチスケール MD による V1ATPase の回転機構の解明**
Rotation mechanism of V1-ATPase studied by multi-scale MD simulation
Yuta Isaka[1], Yuuichi Kokabu[1], Toru Ekimoto[1], Takeshi Murata[2,3], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. of Med Life Sci., Yokohama City Univ.*, [2]*Fac. of Sci., Chiba Univ.*, [3]*JST, PRESTO*)

1Pos102 **アクチン-ミオシン結合における水の役割の理論解析**
A theoretical analysis on water roles in actin-myosin binding
Hiraku Oshima, Tomohiko Hayashi, Masahiro Kinoshita (*Inst. Adv. Energ., Kyoto Univ.*)

1Pos103 **タンデム型エフェクターによる T3SA 内エフェクター分泌機序の評価**
Analysis of Type 3 secretion mechanism by "tandem-connected effectors"
Kyoko Momiyama, Takashi Ohgita, Kentaro Kogure (*Kyoto Pharm. Univ.*)

1Pos104 **蛍光性 NTA-Ni の合成とフォトクロミック分子で修飾した有志分裂キネシン Eg5 への蛍光ラベリング**
Synthesis of fluorescent NTA-Ni and its application to the fluorescent labeling of mitotic kinesin Eg5 modified with photochromic molecule
Yuki Tamura, Kei Sadakane, Ryoma Yamamoto, Kentaro Saito, Shinsaku Maruta (*Division of Bioinformatics, Graduate School of Engineering, Soka University*)

1Pos105 **構成論的手法による真核生物鞭毛運動の機能再構築**
Reconstruction of flagellar motility by the bottom-up strategy
Junya Kirima[1], Misaki Shiraga[2], Hiroaki Kojima[3], **Kazuhiro Oiwa**[1,3,4] ([1]*Grad. Sch. Univ. Hyogo*, [2]*F. Sci., Univ. Hyogo*, [3]*NICT, Adv. ICT. Res. Inst.*, [4]*CREST, Biodynamics*)

1Pos106 **アゾベンゼン誘導体を利用したキネシン二量体化の光制御**
Photo-regulation of kinesin dimerization using bifunctional azobenzene derivatives
Haruka Fujio[1], Yasunobu Sugimoto[2], Kazunori Kondo[3], Shinsaku Maruta[1,3] ([1]*Div. of Bioinfo, Gra. Sch. of Eng., Univ. of Soka*, [2]*Nagoya Univ., SR Research Center*, [3]*Dep. of Bioinfo., Fac. of Eng., Univ. of Soka*)

1Pos107 **ATP 合成酵素の回転運動の幾何学的位相モデル**
Geometric-phase model for the rotary motion of ATP synthase
Tomohiro Yanao (*Grad. Sch. Sci. & Eng., Waseda Univ.*)

1Pos108 ***Mycoplasma mobile*** **の滑走に必須な Gli123 タンパク質の構造解析**
Structure analyses of Gli123 protein, essential for gliding of *Mycoplasma mobile*
Daiki Matsuike, Yuhei O. Tahara, Tasuku Hamaguchi, Makoto Miyata (*Grad. Sch. Sci., Osaka City Univ.*)

1Pos109 **カタユウレイボヤ精子鞭毛における内腕ダイニンのキャラクタリゼーション**
Characterization of inner arm dyneins from sperm flagella in *Ciona intestinalis*
Osamu Kutomi[1], Katsutoshi Mizuno[1,2], Keiko Hirose[3], Lixy Yamada[4], Hitoshi Sawada[4], Kogiku Shiba[1], Kazuo Inaba[1] ([1]*Shimoda Mar. Res. Ctr.*, [2]*Dartmouth College*, [3]*Biomed. Res. Inst., AIST*, [4]*Sugashima Marine Biological Lab., Grad. Sch. Sci., Nagoya Univ.*)

1Pos110 **Dynactin is a biphasic regulator to promote and inhibit dynein motility**
Takuya Kobayashi[1], Takuya Miyashita[1], Hatsuha Kajita[1], Kei Saito[1], Takashi Murayama[2], Yoko Y. Toyoshima[1] ([1]*Grad. Sch. Arts and Sci., Univ. of Tokyo*, [2]*Sch. Med., Juntendo Univ.*)

1Pos111 **カビ由来セルラーゼ Trichoderma reesei Cel6A および Cel7A の逐次運動の１分子蛍光観察**
Single-molecule fluorescence imaging analysis of processive movement of fungal cellulases Trichoderma reesei Cel6A and Cel7A
Tomoyuki Tasaki[1], Akihiko Nakamura[2,3], Daiki Ishiwata[2,3], Hiroyuki Noji[1], Ryota Iino[2,3] ([1]*School of Engineering, The University of Tokyo*, [2]*Okazaki Inst. Integ. Biosci., NINS*, [3]*School of Physical Sciences, SOKENDAI*)

1Pos112 **High-speed angle-resolved imaging of domain motion of the catalytic β subunit of F_1-ATPase**
Sawako Enoki[1], Ryota Iino[2], Yoshihiro Minagawa[1], Yamato Niitani[3], Michio Tomishige[3], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem, Grad. Sch. Eng.*, [2]*Okazaki Inst. Integ. BioSci., NINS*, [3]*Dept. Appl. Phys, Grad. Sch. Eng.*)

1Pos113 **微小管モータータンパク質による微小管構造の識別機構の解析**
In vitro analysis of smart motor hypothesis
Tomohiro Shima[1,2], Yasushi Okada[2] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*QBiC, Riken*)

1Pos114 **バクテリアべん毛モーターにおける二軌道回転**
Two orbital rotation of bacterial flagellar motor
Yoichiro Sawano[1], Yuichi Inoue[1], Akihiko Ishijima[2] ([1]*IMRAM, Tohoku University*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

1Pos115 **F_1-ATPase の回転拡散係数に関する理論研究**
Theoretical study of rotational diffusion for F_1-ATPase
Ryota Shinagawa, Kazuo Sasaki (*Dept. Applied Physics, Tohoku Univ.*)

1Pos116 **べん毛モータートルク発生に関わる構成蛋白質 FliG の多量体の検出**
Identification of multimeric forms of FliG, a flagellar motor component for torque generation
Tohru Umemura[1], Yoshiyuki Sowa[1,3], Ikuro Kawagishi[1,2,3] ([1]*Dept. Bio. Fac. Bio., Hosei Univ.*, [2]*Dept. Eng. Fac. Bio., Hosei Univ.*, [3]*Micro. Nanotec., Hosei Univ.*)

1Pos117 **非平衡効果が引き起こす F_1 分子モーターの非アレニウス型温度依存性**
Non-Arrhenius Type Temperature Dependence of F_1 Molecular Motor Induced by Non-equilibrium Effects
Yuji Tamiya[1,2], Chun-Biu Li[1,2], Rikiya Watanabe[3], Hiroyuki Noji[3], Tamiki Komatsuzaki[1,2] ([1]*Grad. Sch. Sci., Hokkaido Univ.*, [2]*Research Institute for Electronic Science, Hokkaido Univ.*, [3]*Faculty of Engineering, Univ. Tokyo*)

1Pos118 **走化性シグナル CheY-P を結合したべん毛基部体スイッチ複合体の構造**
Structure of the flagellar basal body swich complex with chemotaxis signal protein CheY-P
Tomoko Miyata[1,2], Takayuki Kato[2], Yusuke V. Morimoto[1,2], Hideyuki Matsunami[3], Keiichi Namba[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Trans-Membrane Trafficking Unit, OIST*)

1Pos119 **好熱菌 F_1 の βE190 残基は構造を安定化することにより、F_1 の高効率エネルギー変換に貢献する**
TF_1 βE190 residue contributes to high energy transduction efficiency by stabilizing the structure of the enzyme
Mana Tanaka[1], Tomohiro Kawakami[1], Yohei Nakayama[1], Shoichi Toyabe[3], Hiroshi Ueno[2], Seishi Kudo[3], Eiro Muneyuki[1] ([1]*Dept. Phys., Faculty of Science and Engineering, Chuo Univ.*, [2]*Dept. Appl. Chem., Sch. Eng., Tokyo Univ.*, [3]*Dept. Appl. Phys., Sch. Eng., Tohoku Univ.*)

1Pos120 **2 種類のイオンで駆動する細菌べん毛モーターのエネルギー変換機構の解明**
Analysis of bacterial flagellar rotation driven by dual ion
Kenta Arai[1], Yuka Takahashi[2], Masahiro Ito[2], Yoshiyuki Sowa[1] ([1]*Hosei Univ.*, [2]*Toyo Univ.*)

細胞生物学 / Cell biology

1Pos121 **The Growth Rate of Vibrio alginolyticus Polar Flagellum Decays Exponentially with Flagellar Length**
Mei-Ting Chen (*National Central University*)

1Pos122 **マスト細胞の脱顆粒におけるミトコンドリアカルシウムユニポーターの役割**
Role of mitochondrial calcium uniporter in mast cell degranulation
Tadahide Furuno, Narumi Shinkai, Masanari Ishikawa, Yoshikazu Inoh, Mamoru Nakanishi (*Sch. Pharm., Aichi Gakuin Univ.*)

1Pos123 **1 分子観察によるガングリオシドのダイマー形成機構の解明**
Unraveling of mechanisms of ganglioside dimer formation as revealed by single-molecule imaging
Kenichi Suzuki[1], Hiromune Ando[1,2], Naoko Komura[1,2], Ayano Yamazaki[2], Hideharu Ishida[2], Koichi Furukawa[3], Kenichi Morigaki[4], Akihiro Kusumi[1], Makoto Kiso[1,2] ([1]*iCeMS, Kyoto Univ.*, [2]*Dpt. Appl. Biol. Sci., Gifu Univ.*, [3]*Dpt. Biochem., Nagoya Univ.*, [4]*Res. Ctr. Env. Genomics, Kobe Univ.*)

1Pos124 ミトコンドリア輸送・ATP 産生の同時測定と相関評価
Simultaneous imaging and correlation analysis of mitochondrial trafficking and its ATP production
Rika Suzuki, Kohji Hotta, Kotaro Oka (*Keio University, BioPhysics and NeuroInformatics Lab.*)

1Pos125 好中球様 HL-60 細胞の繰返し基質伸展刺激下での運動方向決定
Directional migration of neutrophil-like HL-60 cells by cyclic substratum stretching
Chika Okimura[1], Kazuki Ueda[1], Yuichi Sakumura[2,3], Yoshiaki Iwadate[1] ([1]*Facult., Sci., Yamaguchi Univ.*, [2]*Sch. Info. Sci. Tech., Aichi Pref. Univ.*, [3]*Grad. Sch. Biol. Sci., Nara Inst. Sci. Tech.*)

1Pos126 構造的微小管結合蛋白質を介した微小管とアクチンフィラメント間の相互作用
Interaction between microtubules and actin filaments via structural microtubule-associated proteins
Miyuki Shiga[1], Shouma Saitou[1], Yurika Hashi[1], Kazushi Matui[1], Susumu Kotani[2], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Sustain. Environ. Eng., Muroran Inst.*, [2]*Kanagawa University*)

1Pos127 心臓組織片による自律拍動の同期化メカニズム
Synchronization process of cardiac tissue fragments
Tomonori Takahashi[1], Kentaro Ishida[1], Tomoyuki Kaneko[2], Toshiyuki Mitsui[1] ([1]*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*, [2]*Bioscience and applied chemistry, Hosei Univ.*)

1Pos128 Mycoplasma mobile のストール力
Stall force of Mycoplasma mobile
Masaki Mizutani[1], Yoshiaki Kinosita[2], Takayuki Nishizaka[2], Makoto Miyata[1] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Fac. of Sci., Gakushuin Univ.*)

1Pos129 定常状態の大腸菌における走化性受容体クラスターの協同性と細胞内シグナル伝達の関係
Relationship between cooperativity in receptor array and intracellular signaling under steady-state of *Escherichia coli*
Hajime Fukuoka[1], Yong-Suk Che[1], Tomoko Horigome[2], Yuichi Inoue[3], Hiroto Takahashi[3], Akihiko Ishijima[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Life Sci, Tohoku Univ.*, [3]*IMRAM, Tohoku Univ.*)

1Pos130 細胞性粘菌における細胞集団の組織的運動の 3 次元解析
3D analysis of collective cell migration in Dictyostelium
Hidenori Hashimura[1], Masato Yasui[2], Kei Inouye[3], Masahiro Ueda[1] ([1]*Department of Biological Sciences, Graduate School of Science, Osaka University*, [2]*RIKIEN QBiC*, [3]*Graduate School of Science, Kyoto University*)

1Pos131 細胞性粘菌の高圧処理からの回復過程で糸状仮足突出が増加する
Protrusion of filopodia increase in the recovery process from the high-pressure treatment of *Dictyostelium* cells
Yuki Gomibuchi[1], Masayoshi Nishiyama[2], Kaoru Katoh[3], Taro Uyeda[3], Takeyuki Wakabayashi[1] ([1]*Teikyo Univ.*, [2]*Kyoto Univ.*, [3]*AIST*)

1Pos132 損傷時の脳がん幹細胞の膜タンパク質の運動解析
Molecular dynamics of brain tumor stem cell induced by specific cell damage
Morito Sakuma, Sayaka Kita, Hideo Higuchi (*Grad. Sch. Sci., Univ. Tokyo*)

1Pos133 **Local intracellular temperature increase mediates stress granule formation**
Beini Shi[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Graduate School of Pharmaceutical Sciences, the University of Tokyo*, [2]*JST-PRESTO*)

1Pos134 蛍光イメージングによる単一ミトコンドリアマトリクス内 ATP 濃度測定
Single mitochondrion imaging of ATP concentration changes in the matrix
Saki Yamashita[1], Takahiro Shibata[1], Kotoe Hirusaki[1], Kaoru Katoh[2], Yoshihiro Ohta[1] ([1]*Grad. Sch. Life Sci. & Bio Tech., TUAT*, [2]*AIST*)

1Pos135 らせん細菌スピロヘータの調和された回転運動
Coordinated cell rotation in the spirochete Leptospira
Kyosuke Takabe, Md. Shafiqul Islam, Shoichi Toyabe, Seishi Kudo, Shuichi Nakamura (*Grad. Sch. Engineering., Univ. Tohoku*)

1Pos136 **Computer simulation of the periodic dynamics of actomyosin foci in C. elegans embryos**
Masashi Fujita[1], Shuichi Onami[1,2] ([1]*RIKEN QBiC*, [2]*NBDC, JST*)

1Pos137 線維芽細胞を介した心筋細胞集団の同期
Synchronization between large clusters of cardiomyocytes through fibroblasts
Shota Miyakoshi[1], Toshiyuki Mitsui[2], Tomoyuki Kaneko[1] ([1]*LaRC, Grad. Sci. Eng., Hosei Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Univ.*)

1Pos138 多細胞の協同運動における力学量の計測と制御
Measurements and control of cellular mechanical factors during collective cell migration
Takeomi Mizutani, Hisashi Haga, Kazushige Kawabata (*Faculty of Advanced Life Sciences, Hokkaido University*)

1Pos139 海洋性ビブリオ菌のべん毛形成を制御する DnaJ ファミリータンパク質 SflA の構造機能解析
Structural and functional analysis of the DnaJ family protein SflA, that is involved in regulation of flagellation in Vibrio alginolyticus
Satoshi Inaba[1], Takehiko Nishigaki[1], Shoji Nishikawa[2], Mayuko Sakuma[1], Seiji Kojima[1], Katsumi Imada[2], Michio Homma[1] ([1]*Div. Biol. Sci. Grad. Sch. Sci. Nagoya Univ.*, [2]*Grad. Sch. Sci. Osaka Univ.*)

1Pos140 微小管を内包した巨大リポソームは架橋・枝分かれ因子無しでも自発的に多角形的な形状になる
Giant liposomes containing microtubules spontaneously develop into polygonal shape in the absence of any crosslinking or branching factor
Masahito Hayashi, Kingo Takiguchi (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos141 細胞における多種分子活性化の同時計測
Simultaneous detection of the activation of signaling proteins in carcinoma cell lines
Hiraku Miyagi[1], Michio Hiroshima[2], Atsushi Mochizuki[1], Yasushi Sako[1] ([1]*RIKEN*, [2]*RIKEN QBiC*)

1Pos142 **心筋細胞に対する力学的刺激の与える影響**
Influence of mechanical stimulus on heart cell aggregates
Shin Arai, Ayaha Tsuyuki, Takashi Nakamura, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

生体膜･人工膜：構造･特性 / Biological & Artificial membrane: Structure & Property

1Pos143 **^{31}P 固体 NMR によるペプチドホルモングルカゴンのアミロイド線維形成過程での生体膜との相互作用変化の解明**
Aging in interaction of glucagon with DMPC lipid bilayers in the process of fibril formation as revealed by ^{31}P solid-state NMR
Kazumi Haya, Akie Kikuchi, Izuru Kawamura, Akira Naito (*Grad. Sch. Eng.,Yokohama Natl. Univ.*)

1Pos144 **Structure-property of ganglioside GM3/DPPC membranes using coarse-grained molecular dynamics simulation**
Kento Inoue[1], Eiji Yamamoto[1], Daisuke Takaiwa[2], Kenji Yasuoka[2], Masuhiro Mikami[2] ([1]*Grad. Sch. Sci. Tech., Keio Univ.*, [2]*Dept. Mech. Eng., Keio Univ.*)

1Pos145 **逆相法の再検討：ホスファチジルコリン巨大ベシクル形成**
A reverse phase method for the formation of giant vesicles (GVs) of phosphatidylcholine
Kanta Tsumoto, Jin Tabata (*Grad. Sch. Eng., Mie Univ.*)

1Pos146 **化学反応下での脂質二重膜のバディングとコラプス**
Budding and collapse of bilayer membrane induced by chemical reaction
Koh Nakagawa, Hiroshi Noguchi (*Institute for Solid State Physics, University of Tokyo*)

1Pos147 **非対称飽和アシル鎖を有するホスファチジルコリンの温度および圧力誘起二重膜相転移**
Temperature- and pressure-induced bilayer phase transitions of phosphatidylcholines with asymmetric saturated acyl chains
Masaki Goto, Nobutake Tamai, Hitoshi Matsuki (*Institute of Technology and Science, Tokushima University*)

1Pos148 **ホスファチジルエタノールアミンを用いて調製したリポソームの特徴**
Characteristics of liposomes containing phosphatidylethanolamine
Hayato Akizuki, Tomoyuki Kaneko (*LaRC, Grad. Sci. Eng., Hosei Univ.*)

1Pos149 **分子動力学計算によるセラミド/水界面の水和構造解析**
Hydration structure at ceramide/water interface: A molecular dynamics simulation study
Suyong Re, Wataru Nishima, Tahei Tahara, Yuji Sugita (*RIKEN, Wako*)

1Pos150 **短鎖リン脂質の吸着膜およびミセルに対するアデノシンリン酸の親和性に及ぼすリン酸基の影響**
Effect of phosphate groups on affinities of adenosine phosphates to adsorbed film and micelles of short-chain phospholipid
Ayumi Nishimaru, Miki Tanaka, Michio Yamanaka (*Fac. Sci., Kyushu univ.*)

生体膜･人工膜：動態 / Biological & Artificial membrane: Dynamics

1Pos151 **Effect of Osmotic Pressure on Constant Tension-Induced Pore Formation in Lipid Membranes**
Sayed Shibly Ul Alam[1], Mohammad Abu Sayem Karal[1], Masahito Yamazaki[1,2] ([1]*Int. Biosci., Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Electronics, Shizuoka Univ.*)

1Pos152 **抗菌ペプチド・ラクトフェリシンＢが誘起する巨大リポソーム中のポア形成**
Antimicrobial Peptide Lactoferricin B-Induced Pore Formation in Single Giant Unilamellar Vesicles
Md. Moniruzzaman[1], Jahangir Md. Alam[2], Hideo Dohra[3], Masahito Yamazaki[1,2,4] ([1]*Int. Biosci., Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Electronics, Shizuoka Univ.*, [3]*Res. Inst. Green Sci. Tech., Shizuoka University*, [4]*Dept. Phys., Grad. Sch. Sci., Shizuoka Univ.*)

1Pos153 **脂質膜表面上での自己組織化された DNA ナノ構造体の構築**
Construction of self-assembled DNA nanostructures on lipid membrane surface
Masamune Morita[1], Miho Yanagisawa[2], Shogo Hamada[3], Shin-ichiro Nomura[4], Satoshi Murata[4], Masahiro Takinoue[1] ([1]*Interdisciplinary Grad. Sch. Sci. and Eng., Tokyo Tech.*, [2]*Dept. Appl. Phys., Tokyo Univ. Agri. Tech.*, [3]*Kavli Inst., Cornell Univ.*, [4]*Grad. Sch. Eng., Tohoku Univ.*)

1Pos154 **バクテリアを封入したリポソームにおける大腸菌の動態**
Dynamics of Escherichia coli in bacteria-enclosed liposomes
Hazuki Terajima, Ryutaro Isobe, Tomoyuki Kaneko (*LaRC., Dept. Frontier Biosci., Hosei Univ.*)

1Pos155 **リポソーム融合の光マニピュレーション**
Photocontrol of liposome fusion
Yui Suzuki[1], Kazuki Shigyou[1], Ken Nagai[1], Anatoly Zinchenko[2], Tsutomu Hamada[1] ([1]*Sch. of Materials Science, JAIST*, [2]*Grad. Sch. of Environmental Studies, Nagoya Univ.*)

生体膜･人工膜：興奮･チャネル / Biological & Artificial membrane: Excitation & Channels

1Pos156 **Simple method for lipid bilayer formation with simultaneous incorporation of ion channels using gold electrode (2)**
Toru Ide[1,2], Daichi Okuno[2], Takamitsu Kira[1], Minako Hirano[2,3], Hiroaki Yokota[3] ([1]*Grad Schl Natl Sci Tech, Okayama Univ*, [2]*Riken*, [3]*GPI*)

1Pos157 **高速原子間力顕微鏡で見る膜中カリウムチャネル KcsA の動的挙動**
Dynamic behavior of the KcsA potassium channel in membrane observed by high-speed atomic force microscopy
Ayumi Sumino[1,2], Takayuki Uchihashi[3], Daisuke Yamamoto[4], Masayuki Iwamoto[2], Takehisa Dewa[5], Shigetoshi Oiki[2] ([1]*JST/PRESTO*, [2]*Facult. Med. Sci., Univ. Fukui*, [3]*Depart. Phys., Kanazawa Univ.*, [4]*Facult. Sci., Univ. Fukuoka*, [5]*Grad. Sch. Eng., Nagoya Inst. Tech.*)

1Pos158 **ポリセオナミドBチャネルの pH 依存性ゲーティング機構**
pH-dependent gating of the polytheonamide B channel
Yuka Matsuki[1], Masayuki Iwamoto[1], Shigeki Matsunaga[2], Shigetoshi Oiki[1] ([1]*Dept. Mol. Physiol. Biophys., Univ. Fukui Fac. Med. Sci., Univ, Fukui, Japan*, [2]*Lab. Aqua. Nat. Products Chem., Grad. Sch. Agri. Life Sci., Univ. Tokyo, Japan*)

生体膜･人工膜：輸送 / Biological & Artificial membrane: Transport

1Pos159 **細菌べん毛 III 型分泌装置のエネルギー変換メカニズム**
An energy transduction mechanism of the bacterial flagellar type III secretion apparatus
Hiroyuki Terashima[1], Akihiro Kawamoto[2], Chinatsu Tatsumi[1], Tohru Minamino[2], Keiichi Namba[2], Katsumi Imada[1] ([1]*Dep. Macromol. Sci., Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

1Pos160 **Zero-Mode Waveguides sealed with artificial lipid bilayer for the analysis of membrane proteins**
Keisuke Nagao[1], Toshihisa Osaki[2], Ryota Iino[3], Takayuki Uchihashi[4], Hirofumi Shintaku[1], Hidetoshi Kotera[1], Ryuji Yokokawa[1] ([1]*Kyoto University*, [2]*The University of Tokyo*, [3]*Institute for Molecular Science*, [4]*Kanazawa University*)

生体膜･人工膜：信号伝達 / Biological & Artificial membrane: Signal transduction

1Pos161 **PI3K と Ras から成るフィードバック回路による自己組織的な PIP3 局在形成の制御**
Positive feedback loop composed by PI3K and Ras regulates self-organization of PIP3-enriched domain
Seiya Fukushima[1,2], Satomi Matsuoka[2], Masahiro Ueda[1,2] ([1]*Graduate School of Science, Osaka University*, [2]*Riken Quantitative Biology Center*)

神経科学･感覚 / Neuroscience & Sensory systems

1Pos162 **海馬シナプスへの性ホルモンの急性作用：男性ホルモンと女性ホルモンの比較**
Acute effect of estrogen and androgen on hippomcapal synapses
Yasushi Hojo[1,2], Yoshitaka Hasegawa[2,3], Yusuke Hatanaka[3], Bon-chu Chung[2], Suguru Kawato[2,3,4] ([1]*Dept. Biochem., Saitama Med. Univ.*, [2]*JST, Japanese-Taiwanese Cooperative Programme*, [3]*Grad. Sch. of Arts and Sci., Univ. of Tokyo*, [4]*Urology, Juntendo Univ.*)

1Pos163 **アルギン酸ゲルを用いた神経シートの作製技術の開発と機能評価**
Fabrication and functional evaluation of “neuronal sheet” using calcium alginate gel
Hideyuki Terazono, Hyonchol Kim, Fumimasa Nomura, Kenji Yasuda (*Biomed. info., IBB, Tokyo Med. Dent. Univ.*)

1Pos164 **β3 チューブリン変異による軸索伸長の異常を、キネシンのサプレッサー変異で回復する**
Reversal of axon growth defects in CFEOM3 by suppressor mutation in the kinesin-microtubule interface
Itsushi Minoura[1], Hiroko Takazaki[1,2], Rie Ayukawa[1], You Hachikubo[1], Yoshihiko Yamakita[1,3], Seiichi Uchimura[1], Chihiro Yoshida[1], Tomomi Shimogori[1], Tomonobu Hida[1], Hiroyuki Kamiguchi[1], Etsuko Muto[1] ([1]*BSI, RIKEN*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Sch. Med. Nagoya Univ.*)

神経回路･情報処理 / Neuronal circuit & Information processing

1Pos165 **培養神経回路網における機能的結合のグラフ構造**
The graph structure of functional connections in a cultured neuronal network
Nanami Hirata, Hidekatsu Ito, Wataru Minoshima, Suguru Kudoh (*Kwansei Gakuin Univ*)

1Pos166 **リアノジン受容体を介した神経細胞の温度感受性カルシウム放出**
Thermosensitive Ca2+ burst in rat hippocampal neurons through ryanodine receptors
Yuki Kawamura[1], Kotaro Oyama[1,2], Hideki Itoh[1,3], Vadim Zeeb[4], Madoka Suzuki[5,6], Shin’ichi Ishiwata[1,5,6] ([1]*Sch. Adv. Sci. Eng., Waseda Univ.*, [2]*Dept. Cell Physiol., The Jikei Univ. Sch. Med.*, [3]*Inst. Med. Biol., A*STAR, Singapore*, [4]*Inst. Theoret. Exp. Biophys., Rus. Acad. Sci.*, [5]*WASEDA Biosci Res Inst Singapore (WABIOS)*, [6]*Org Univ Res Initiatives, Waseda Univ*)

行動 / Behavior

1Pos167 **滑走細菌 *Flavobacterium johnsoniae* のレール状滑走装置**
Rail-like structure in gliding machinery of the gliding bacterium *Flavobacterium johnsoniae*
Satoshi Shibata[1], Akihiro Kawamoto[2], Takayuki Katou[2], Keiichi Namba[2], Koji Nakayama[1] ([1]*Graduate Sch. of Biomedical Science, Nagasaki Univ*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

光生物：視覚･光受容 / Photobiology: Vision & Photoreception

1Pos168 ***In-situ* 光照射固体 NMR による D96N-bR 変異体の光中間体と光反応経路の解析**
Characterization of photo-intermediates and photo reaction pathway of D96N-bR mutant by *in-situ* photo-irradiation solid-state NMR
Yuto Otani[1], Arisu Shigeta[1], Yoshiteru Makino[1], Ryota Miyasa[1], Izuru Kawamura[1], Takashi Okitsu[2], Akimori Wada[2], Satoru Tuzi[3], Akira Naito[1] ([1]*Grad. Sch. Eng, Yokohama Natl Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Univ. of Hyogo*)

1Pos169 **局所的な照明により制御される IV 型線毛の収縮**
Retraction of Type IV pili controlled by local light gradient
Daisuke Nakane, Takayuki Nishizaka (*Dept. of Phys., Gakushuin Univ.*)

1Pos170 ***Harorubrum* 族の新種から見つかったアーキロドプシンの大腸菌での発現**
Archaerhodopsin found in *Harorubrum* sp. ejinoor was functionally expressed in *Escherichia coli*
luomeng Chao[1], Xiong Geng[1], Gang Dai[2], Takashi Kikukawa[3], Tatsuo Iwasa[1] ([1]*1Div. Eng. Composite Funct., Muroran Ins. Technol., Japan*, [2]*Coll. Chem. Environ. Sci., Inner Mongolia Normal Univ., China*, [3]*Grad. Sch. Life. Sci., Hokkaido Univ., Japan*)

1Pos171 **ニワトリクリプトクロム 4 の光反応の特性解析**
Characterization of photoreaction property of chicken cryptochrome4
Hiromasa Mitsui, Keiko Okano, Toshiyuki Okano (*Dept. Eng. and Biosci., Grad. Sch. Adv. Sci. and Eng., Waseda Univ.*)

1Pos172 **YFP および mCherry と融合させた光制御型 bZIP タンパク質（Photozipper）の解析**
Analyses of a light-regulated bZIP protein, Photozipper, fused with YFP and mCherry
Keigo Furuya, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

1Pos173 **水溶液中のオキシルシフェリン光ルミネッセンス過程**
Analysis of Photoluminescence Pathways of Firefly Oxyluciferin in Aqueous Solution
Miyabi Hiyama[1], Toshimitsu Mochizuki[2], Hidefumi Akiyama[1], Nobuaki Koga[3] ([1]*ISSP, Univ. Tokyo*, [2]*AIST*, [3]*Grad. Sch. Info. Sci., Nagoya Univ.*)

1Pos174 **PYP Binding Protein の Rc-PYP との複合体形成における N 末端領域の役割**
The role of N-terminal region of PYP binding protein at the formation of complex with Rc-PYP
Tomoyuki Yasumuro, Yoichi Yamazaki, Mikio Kataoka, Hironari Kamikubo (*Grad. Sch. Mat. Sci., NAIST*)

1Pos175 **シアノバクテリアに由来する新規クロライドポンプ型ロドプシンは一残基置換によってプロトンポンプへ変換される**
A novel chloride–pumping rhodopsin from cyanobacterium converts to proton pump with a single amino acid replacement
Takatoshi Hasemi, Takashi Kikukawa, Masakatsu Kamiya, Tomoyasu Aizawa, Naoki Kamo, Makoto Demura (*Grad. Sch. Life Sci., Hokkaido Univ.*)

1Pos176 **ナトリウムポンプ型ロドプシン KR2 を基にしたカリウムポンプの創成**
Engineering K^+ pumping rhodopsin from KR2
Masae Konno[1], Keiichi Inoue[1,2], Hideaki Kato[3], Osamu Nureki[3], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*PRESTO, JST*, [3]*Grad. Sch. Sci., Univ. of Tokyo*)

1Pos177 **ファラオニスハロロドプシンの細胞質側チャネルに位置する重要残基の探索**
Functionally important residues in the cytoplasmic half channel of light-driven Cl^- pump *Natronomonas pharaonis* halorhodopsin
Anna Shimosaka, Takashi Kikukawa, Masakatsu Kamiya, Tomoyasu Aizawa, Naoki Kamo, Makoto Demura (*Grad. Sch. Life Sci., Univ. Hokkaido*)

光生物：光合成 / Photobiology: Photosynthesis

1Pos178 ***Tch. tepidum* 由来 LH1-RC の異常吸収挙動と熱耐性の解明に向けて**
Toward elucidating the unusual absorption behavior and enhanced thermostability of the LH1-RC complex from *Tch. tepidum*
T. Kawakami[1], L.-J. Yu[1,2], Yukihiro Kimura[3], S. Otomo[1] ([1]*Ibaraki Univ.*, [2]*Present address: Grad. Sch. Biol., Okayama Univ.*, [3]*Grad. Sch. Agri. Sci., Kobe Univ.*)

1Pos179 **好熱性紅色光合成細菌由来 LH1-RC 複合体の金属結合サイトおよび金属—タンパク質間相互作用の観測**
Monitoring of metal-binding sites and metal-protein interactions in the LH1-RC complex from thermophilic purple photosynthetic bacteria
Yuki Yura[1], Yukihiro Kimura[1], Yusuke Hayashi[1], Li Yong[1], Moe Onoda[1], Seiu Otomo[2], Takashi Ohno[1] ([1]*Grad. Sch. Agri. Sci., Kobe Univ.*, [2]*Fac. Sci., Ibaraki Univ.*)

1Pos180 **多孔性シリカ粒子のナノ空間に埋め込まれたヘリオバクテリア反応中心コアタンパクの安定性と分光学的特性**
Stability and spectroscopic characterization of heliobacterial reaction center core protein incorporated into nanoporous silica particles
Hirozo Oh-oka[1], Tomoyasu Noji[2], Chihiro Azai[3], Risa Mutoh[4], Genji Kurisu[4], Shigeru Itoh[5] ([1]*Department of Biological Sciences, Graduate School of Science, Osaka University*, [2]*The OCU Advanced Research Institute for Natural Science and Technology, Osaka City University*, [3]*Department of Bioinformatics, College of Life Science, Ritsumeikan University*, [4]*Institute for Protein Research, Osaka University*, [5]*Center for Gene Research, Nagoya University*)

1Pos181 **極低温顕微鏡を用いた緑化途上トウモロコシ生葉の光合成タンパク質前駆体の空間分布の測定**
Cryogenic microscope observations of photosynthetic proteins under the assembly process in greening etiolated Zea mays leaves
Tomofumi Chiba, Hiroshi Fukumura, Yutaka Shibata (*Grad. Sch. Sci., Univ. Tohoku*)

1Pos182 **高酸化状態のマンガンクラスターにおける Ca^{2+}の影響**
Influence of Ca^{2+} on spin structure of Mn cluster in high oxidation state
Takahiro Sakai[1], Hiroki Nagashima[1], Hitomi Matsushita[2], Ko Furukawa[3], Hiroyuki Mino[1] ([1]*Grad. Sch. Sci., Univ. Nagoya*, [2]*Sch. Sci., Univ. Nagoya*, [3]*Fac., Sci., Univ. Niigata*)

1Pos183 **光合成水分解反応における S0→S1 遷移の時間分解赤外分光解析**
Time-resolved infrared analysis of the S0-to-S1 transition of photosynthetic water oxidation
Tatsuki Shimizu, Ryo Nagao, Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

放射線生物学·活性酸素 / Radiobiology & Active oxygen

1Pos184 **FRET を用いた放射線誘発 DNA 脱塩基部位の局在性評価**
Localization estimation of abasic (AP) sites in DNA induced by ionizing radiation
Ken Akamatsu, Naoya Shikazono (*Japan Atomic Energy Agency*)

1Pos185 **酸化ストレスはビタミン D によって誘発される骨髄細胞分化を調整する**
Oxidative Stress Modulates Vitamin D-induced Myeloid Cell Differentiation
Hiroaki Tanaka[1], Hiroyuki Kato[1], Omi Nawa[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Mater. & Human Env. Sci., Shonan Inst. of Tech.*)

生命の起源·進化 / Origin of life & Evolution

1Pos186 **大腸菌とマイクロデバイスのハイブリッド型人工細胞の効率改善に向けた条件検討**
The suitable condition for the hybrid artificial cell system based on *E. coli* and the ALBiC device
Yoshiki Moriizumi[1], Kazuhito V. Tabata[1,3], Rikiya Watanabe[1,3], Hiroyuki Noji[1,2] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ Tokyo*, [2]*CREST, JST*, [3]*PRESTO, JST*)

1Pos187 **鋳型複製する高分子系の数理モデルにおける配列情報の選択**
Sequence selection in mathematical model of template replicating polymer system
Yoshiya Matsubara, Kunihiko Kaneko (*Department of Basic Science, The University of Tokyo*)

ゲノム生物：ゲノム構造 / Genome biology: Genome structure

1Pos188 **Visualization of chromatin dynamics and domains in live mammalian cells**
Tadasu Nozaki[1,2], Sachiko Tamura[1], Ryosuke Imai[1], Tomomi Tani[3], Masaru Tomita[2], Takeharu Nagai[4], Yasushi Okada[5], Kazuhiro Maeshima[1] ([1]*Natl. Inst. Genet.*, [2]*Inst. Adv. Biosci., Keio Univ.*, [3]*Marine Biological Laboratory*, [4]*ISIR, Osaka Univ.*, [5]*QBiC, RIKEN*)

バイオインフォマティクス：ゲノム構造 / Bioinformatics: Structural genomics

1Pos189 **フォールド構造に基づく膜タンパク質の分類**
Classification of transmembrane proteins based on the fold structure
Tsukasa Ueno[1], Masami Ikeda[1,2], Makiko Suwa[1,2] ([1]*Biol. Sci., Grad. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Chem. Biol. Sci., Sci. Eng., Aoyama Gakuin Univ.*)

1Pos190 **複合体モデリングにより発見された新規相互作用面と病気関連変異の関係**
Relationship between protein-protein interaction interface predicted by complex modeling and disease-related amino acid variants
Toshiyuki Tsuji, Takao Yoda, Tsuyoshi Shirai (*Nagahama Institute of Bio-Technology and Science*)

1Pos191 **ボツリヌス菌の大型プロジェニター毒素複合体のダイナミクス**
Dynamics of the Large Progenitor Toxin Complex of Clostridium botulinum
Yosuke Kondo[1], Tomonori Suzuki[2], Yeondae Kwon[3], Satoru Miyazaki[4] ([1]*Grad. Sch. Pharm., Tokyo Univ. Sci.*, [2]*Nut. Sci. Food Saf., Tokyo Univ. Agri.*, [3]*Grad. Sch. Agri. Life Sci., Univ. Tokyo*, [4]*Fac. Pharm., Tokyo Univ. Sci.*)

1Pos192 **蛋白質複合体における構造変化のデータベース解析**
Database analysis of structural changes in protein complexes
Ryotaro Koike, Motonori Ota (*Grad. Sch. of Info. Sci., Nagoya Univ.*)

バイオインフォマティクス：ゲノム機能 / Bioinformatics: Functional genomics

1Pos193 **蛋白質の配列進化における最小作用原理とその変異体解析への応用**
Application of the principle of least action to protein sequence evolution for quantifying the effect of mutations
Motoi Taniguchi (*Dept. Biol. Sci., Grad. Sch. Sci., Osaka Univ.*)

バイオインフォマティクス：ゲノム比較 / Bioinformatics: Comparative genomics

1Pos194 **遺伝子発現データと染色体構造データに基づく真核生物の染色体上における遺伝子発現相関の解析**
Analysis of correlations between gene expressions and chromosome structures of eukaryotes based on publicly available data sets
Rei Tanikado[1], Akinori Awazu[1,2], Hiraku Nishimori[1,2] ([1]*Dept. of Mathematical and Life Sci., Univ. Hiroshima*, [2]*Research center for the Mathematics on chromatin live Dynamics (RcMcD)*)

数理生物 / Mathematical biology

1Pos195 **Eddy current flow of probability in stochastic gene expression dynamics in eukaryotes**
Bhaswati Bhattacharyya, Masaki Sasai (*Sasai group, Department of Computational Science and Engineering, Graduate School of Engineering*)

1Pos196 **細胞核の変形運動が核内クロマチン配置に及ぼす効果**
Nuclear deformation dynamics induced hetero- and eu-chromatin positioning
Akinori Awazu[1,2] ([1]*Department of Mathematical and Biosciences, Hiroshima University*, [2]*Research Center for Mathematics on Chromatin live Dynamics, Hiroshima University*)

1Pos197 **適応度と情報に関するゆらぎ定理**
Fluctuation Relations for Fitness and Information
Tetsuya Kobayashi, Yuki Sughiyama (*Institute of Industrial Sciene, University of Tokyo*)

1Pos198 **細胞の自発運動ダイナミクスの種間共通性**
Common aspects of spontaneous cell migration dynamics in different species
Hiroaki Takagi (*Dept. Phys., Nara Med. Univ.*)

1Pos199 **A stochastic simulation study on circadian oscillation and ATPase activity of KaiC hexamer**
Sumita Das, Shota Hashimoto, Tomoki P. Terada, Masaki Sasai (*Department of Computational Science and Engineering, Nagoya University*)

非平衡･生体リズム / Nonequilibrium state & Biological rhythm

1Pos200 **Dynamical model of chromosome synapsis formation during meiosis in Eukaryotes**
K. Takamiya[1], K. Yamamoto[1], Hiraku Nishimori[1,2], A. Awazu[1,2] ([1]*1Dept. Mathematics and Life sciences, Grad. Sch. Sci, Univ. Hiroshima*, [2]*Research Center Math. Chromatin Live Dynamics (RcMcD), Univ. Hiroshima*)

1Pos201 **パルス密度制御による小胞型非平衡開放系リアクタ**
Vesicular nonequilibrium open reactor regulated by pulse-density modulation
Haruka Sugiura[1], Manami Ito[1], Hiroyuki Kitahata[2], Yoshihito Mori[3], **Masahiro Takinoue**[1,4] ([1]*Dept. Comput. Intell. Syst. Sci., Tokyo Tech*, [2]*Dept. Phys., Chiba Univ.*, [3]*Dept. Chem., Ochanomizu Univ.*, [4]*PRESTO, JST*)

1Pos202 **キネシン駆動微小管の非平衡パターン形成**
Pattern formation of microtubules driven by kinesin
Sakurako Tanida[1], Ken'ya Furuta[2], Kaori Nishikawa[1], Hiroaki Kojima[2], Masaki Sano[1] ([1]*Grad. Sch. Sci., The Univ. Tokyo*, [2]*NICT*)

計測 / Measurements

1Pos203 **高速走査型イオン伝導顕微鏡の開発**
Development of high-speed scanning ion conductance microscopy
Shinji Watanabe[1], Toshio Ando[1,2,3,4] ([1]*Bio-AFM, Kanazawa Univ.,*, [2]*Fac. of Math. & Phys., Kanazawa Univ.*, [3]*Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [4]*JST-CREST*)

1Pos204 **Target imaging droplet sorting system: a shape identification method for recognition and sort target droplet with cell in real time**
Mathias Girault[1], Hyonchol Kim[1,2], Kenji Matsuura[1], Masao Odaka[1,2], Hideyuki Terazono[1,2], Akihiro Hattori[1], Kenji Yasuda[1,2] ([1]*KAST*, [2]*TMDU*)

1Pos205 **金ナノ粒子を用いた 1 細胞局所加熱による細胞内温度の研究**
Investigation of intracellular temperature using local heating of a single cell with gold nanoparticles
Takaaki Honda[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharma., Univ. Tokyo*, [2]*JST, PRESTO*)

1Pos206 **レンズドファイバを用いた内視鏡型蛍光相関分光装置の開発**
Development of an endoscopic fluorescence correlation spectroscopy using a lensed fiber
Johtaro Yamamoto, Masataka Kinjo (*Graduate School of Life Science, Hokkaido University*)

1Pos207 **アドレナリンによる培養神経細胞軸索輸送活動度の増加がフロー解析法により定量化された**
Flow analysis revealed the activity increase of axonal transport of cultured neurons by Adrenaline
Takashi Katakura, Risa Isonaka, Tadashi Kawakami (*Dept. Physiol., Kitasato Univ. Sch. Med.*)

バイオイメージング / Bioimaging

1Pos208 **細胞内小器官選択的な蛍光分子温度計**
Organelle-targeting molecular fluorescent thermometers for living cells
Madoka Suzuki[1,2], Satoshi Arai[1], Young-Tae Chang[3] ([1]*WASEDA Biosci Res Inst Singapore (WABIOS)*, [2]*Org Univ Res Initiatives, Waseda Univ*, [3]*Dept Chem, Natl Univ Singapore (NUS)*)

1Pos209 **ストレプトリジン O による膜孔形成の高速 AFM 観察**
High-speed AFM Observation of Membrane Pore Formation by Streptolysin O
Hirotaka Ariyama[1], Noriyuki Kodera[1], Toshio Ando[1,2] ([1]*Bio-AFM Frontier Research Center, Kanazawa Univ.*, [2]*Dept. Phys., Kanazawa Univ.*)

1Pos210 **Development of a novel user-friendly system for image processing of electron micrographs by integrating web browser and PIONE with Eos**
Takafumi Tsukamoto, Takuo Yasunaga (*Kyushu Institute of Technology*)

1Pos211 **X 線自由電子レーザーを用いた低温コヒーレント回折イメージングによる酵母細胞核の三次元構造解析**
Three-dimensional structure of yeast nucleus visualized by cryogenic coherent diffraction imaging using X-ray free-electron laser
Yuki Sekiguchi[1,2], Amane Kobayashi[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Yuichi Ichikawa[3], Hitoshi Kurumizaka[3], Mitsuhiro Shimizu[4], Masaki Yamamoto[2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*, [3]*Grad. Adv. Sci. Eng., Waseda Univ.*, [4]*Grad. Sci. Eng., Meisei Univ.*)

1Pos212 **幅広い応用が可能な超高輝度マルチカラー発光タンパク質**
Color pallet of super-duper luminescent proteins capable of wide range application
Kazushi Suzuki[1], Yoshiyuki Arai[1,2], Masahiro Nakano[1,2], Takeharu Nagai[1,2] ([1]*Grad. Sch. Engin., Osaka Univ.*, [2]*ISIR, Osaka Univ.*)

1Pos213 **Yeast two hybrid および定量イメージング解析による PDLIM2 相互作用タンパク質の同定**
Identification of PDLIM2 interacting protein using yeast two-hybrid system and quantitative imaging analysis
Chanyoung Shin[1,2], Yuma Ito[1], Kumiko Sakata-Sogawa[1], Takashi Tanaka[2], Makio Tokunaga[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*IMS-RCAI, RIKEN*)

1Pos214 **Measurement of the redox potential in Chlamydomonas flagella using a redox-sensitive fluorescent protein, Oba-Q**
Yuta Nishimaki, Kazunori Sugiura, Toru Hisabori, Ken-ichi Wakabayashi (*Chemical Resources Laboratory, Tokyo Institute of Technology*)

1Pos215 **クロマチン動態制御におけるアクチン関連タンパク質 Arp4 の核内ダイナミクス**
Dynamics of actin-related protein 4 in living cell nucleus for dynamic chromatin regulation
Yuma Ito[1], Hiroshi Kimura[1], Masahiko Harata[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

1Pos216 **蛍光褪色回復法による INO80 クロマチンリモデリング複合体の核内動態解析**
Intranuclear dynamics of INO80 chromatin remodeling complex by fluorescent recovery after photobleaching
Yuma Ito[1], **Tsubasa Isogaki**[1], Hiroshi Kimura[1], Masahiko Harata[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

1Pos217 **高速原子間力顕微鏡により観察された AAA シャペロン p97 の主要 ATPase リングの構造変化**
Conformational changes of the major ATPase domain D2 of the AAA chaperone p97 observed by high-speed atomic force microscopy
Daisuke Yamamoto[1,3], Kentaro Noi[2,3], Ken-ichi Arita-Morioka[2,3], Teru Ogura[2,3] ([1]*Fac. Sci., Fukuoka Univ.*, [2]*IMEG, Kumamoto Univ.*, [3]*CREST, JST*)

1Pos218 **軟 x 線顕微鏡による糸状シアノバクテリアにおける窒素固定の直接観察**
Direct observation of nitrogen fixation in filamentous cyanobacteria by using soft X-ray microscopy
Takahiro Teramoto[1], Masashi Yoshimura[2], Chihiro Azai[3], Kazuki Terauchi[3], Hidetoshi Namba[2], Toshiaki Ohta[2] ([1]*Col. Sci. & Eng, Ritsumeikan Univ.*, [2]*SRCenter, Ritsumeikan Univ.*, [3]*Col. Life Sci. Ritusmeikan Univ.*)

バイオエンジニアリング / Bioengineering

1Pos219 **遠心操作を伴わない細胞を回収するためのマイクロデバイス**
Microfluidic Device for Recovering Cells without Centrifugation
Koji Matsuura, Saori Nishina, Keiji Naruse (*Cardiovascular Physiology, Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama University*)

1Pos220 **芽胞菌に対するマイクロ波照射効果**
Microwave Irradiation Effect to Spore-Forming Bacteria
Ryota Nakama[1], Arata Shiraishi[1], Wataru Nagayoshi[1], Sakura Yoshimoto[2], Shokichi Ohuchi[1,2] ([1]*Dept. Lifesci. & Syst, Eng., Kyushu Inst. Tech.*, [2]*Dept. Biosci. & Bioinform., Kyushu Inst. Tech.*)

1Pos221 **ビオチン化したタンパク質ラングミュア膜上に作成したストレプトアビジン層**
Streptavidin layer formed on the biotinylated Langmuir film of protein
Taiji Furuno (*Dept. Phys., Keio Univ. Sch. Med.*)

1Pos222 **光分解性ゲルによる接着細胞の力学的操作法の検討**
Application of photodegradable hydrogel to mechanical manipulation of adherent cells
Kentaro Iketaki[1], Humiki Yanagawa[2], Ryuzo Kawamura[1], Seiichiro Nakabayashi[1], Toshiyuki Takagi[2], Shinji Sugiura[2], Toshiyuki Kanamori[2], Hiroshi Yoshikawa[1] ([1]*Dept. Chem., Saitama Univ.*, [2]*BRD., AIST.*)

1Pos223 **電極埋め込み型ナノポアの AC ゲート電位による DNA の挙動制御**
Controlling DNA motions with an AC gate voltage applied gate embedded in nanopore
Yuta Kato, Naoto Sakashita, Yoshitaka Tanida, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

1Pos224 **ハニカム構造を有するマイクロゲルネットワークの構築**
Construction of microgel network with honeycomb structure
Satoshi Umeyama, Masayuki Hayakawa, Masamune Morita, Masahiro Takinoue (*Interdisciplinary Graduate school of Science and Engineering*)

1Pos225 **Directed evolution system to generate peptide agonists for G protein-coupled receptors using in vitro translation in water-in-oil droplets**
Takashi Sakurai[1], Ryo Iizuka[1], Yasuyuki Nakamura[2], Jun Ishii[3], Rui Sekine[4], Yoon Dong H.[4], Tetsushi Sekiguchi[5], Akihiko Kondo[2], Shuichi Shoji[4], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Eng., Kobe Univ.*, [3]*Org. of Adv. Sci. and Technol., Kobe Univ.*, [4]*Grad. Sch. of Adv. Sci. and Eng., Waseda Univ.*, [5]*Inst. for Nanosci. and Nanotechnol., Waseda Univ.*)

1Pos226 **分子モータと DNA 回路を内包した巨大リポソームにより構築される分子ロボット**
Molecular robots constructed by entrapping molecular-motors and DNA circuits into giant liposomes
Yusuke Sato[1], Yuichi Hiratsuka[2], Ibuki Kawamata[1], Satoshi Murata[1], Shin-ichiro Nomura[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Sch. Matl. Sci., JAIST*)

1Pos227 **光応答基板：細胞移動研究のためのマテリアルバイオロジー的研究ツール**
Photoactivatable substrates: Material biological tools for cell migration research
Jun Nakanishi[1], Kazuo Yamaguchi[2] ([1]*NIMS*, [2]*Kanagawa University*)

1Pos228 **Novel genetically encoded antibody-based biosensors allowing fluorescence ratio detection of antigens**
Kim Phuong Huynh Nhat, Takayoshi Watanabe, Takahiro Hohsaka (*JAIST*)

その他 / Miscellaneous topics

1Pos229 **ベイズ推定を用いた CTF 補正の自動化**
Automation of CTF correction using the Bayesian estimation
Koji Hisanaga, Takuo Yasunaga (*Kyushu Institute of Technology Graduate School of Computer Science and Systems Engineering Yasunaga Lab*)

1Pos230 **Model studies of bacterial flagellar motor switching in response to CheY-P regulation and motor structural alterations**
Qi Ma[1], Matthew A. B. Baker[2], Fan Bai[1] ([1]*Biodynamic Optical Imaging Center, Peking University, Beijing, China*, [2]*Victor Chang Cardiac Research Institute, Sydney, NSW, Australia*)

第 2 日目（9 月 14 日（月））／Day 2（Sep. 14 Mon.）プロムナード / Promenade

蛋白質：構造 / Protein: Structure

2Pos001 **CryoEM 3D image reconstruction of the flagellar LP ring complex**
Meltem Tatli[1], Tomoko Miyata[1,2], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Riken QBiC*)

2Pos002 **毒性型アミロイドテープ： GM1 クラスター上で形成される新規 Aβ 逆平行／平行混合 β シート構造**
Toxic Amyloid Tape: Novel Mixed Antiparallel/Parallel β-Sheet Structure Formed by Aβ on GM1 Clusters
Yuki Okada[1], Keisuke Ikeda[5], Yoshiaki Yano[1], Masaru Hoshino[1], Yoshio Hayashi[4], Yoshiaki Kiso[3], Hikari Itoh-Watanabe[2], Akira Naito[2], **Katsumi Matsuzaki**[1] ([1]*Grad. Sch. Pharm. Sci. Kyoto Univ.*, [2]*Fac. Eng. Yokohama Nation. Univ.*, [3]*Nagahama Inst. Bio-Sci. Tech.*, [4]*Tokyo Univ. Pharm. Life Sci.*, [5]*Grad. Sch. Med. Pharm. Sci. Univ. Toyama*)

2Pos003 **Hsp90 と ADP の会合における水分子の役割**
Role of water molecules for association of Hsp90 and ADP
Kazutomo Kawaguchi, Hiroaki Saito, Hidemi Nagao (*Inst. Sci. Eng., Kanazawa Univ.*)

2Pos004 **赤外分光法によるインスリンアミロイドの構造規則性の解明**
Revealing the structural rules of insulin amyloid by infrared spectroscopy
Hisayuki Morii[1], Takashi Shimizu[1], Masayuki Nara[2] ([1]*AIST*, [2]*Tokyo Med. Dent. Univ.*)

2Pos005 **タンパク質の形状に内在する機構的性質に関する解析**
Analysis of Mechanism Features Present in Protein Shapes
Keisuke Arikawa (*Fcl. Eng. , Kanagawa Inst. of Tech.*)

2Pos006 **抗体 CDR-H3 ループ領域の立体構造予測**
Accurate ensemble modeling of CDR-H3 loop in antibody
Hiroshi Nishigami[1,2], Gert-Jan Bekker[1], Narutoshi Kamiya[1], Junichi Higo[1], Haruki Nakamura[1] ([1]*Institute for Protein Research, Osaka University*, [2]*Department of Biology, Graduate School of Science, Osaka University*)

2Pos007 **単粒子像解析のための分子同定用 GFP ラベル**
GFP protein labeling for single particle image analysis
Takayuki Kato[1], Naoya Terahara[1], Tomoko Miyata[2], Keiichi Namba[1,2] ([1]*Osaka Univ, Front. Biosci.*, [2]*Riken, Qbic*)

2Pos008 **広範囲に基質特異性を示す L-アミノ酸酸化酵素の構造**
Structure of L-amino acid oxidase with broad substrate specificity
Nanako Ito[1], Tatsuya Kawaguchi[1], Kaho Murakami[2], Takashi Tamura[2], Miwa Yamada[3], Kimiyasu Isobe[3], Kenji Inagaki[2], Katsumi Imada[1] ([1]*Dept. MacroMol., Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Env. & Life Sci., Okayama Univ.*, [3]*Dept. Biol. Chem. & Food Sci., Iwate Univ*)

2Pos009 **Thermococcus kodakaraensis 由来エンドヌクレアーゼ NucS の構造**
Structure prediction and analyses of endonuclease NucS from *Thermococcus kodakaraensis*
Setsu Nakae[1], Atsushi Hijikata[1], Toshiyuki Tsuji[1], Koki Yonezawa[1], Ken-ichi Kouyama[2], Kouta Mayanagi[3], Sonoko Ishino[4], Yoshizumi Ishino[4], Tsuyoshi Shirai[1] ([1]*Fac. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*, [2]*Grad. Sch. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*, [3]*Medical Inst. of Bioregulation, Kyushu Univ.*, [4]*Grad. Sch. Bioresource and Bioenviron. Sci., Kyushu Univ.*)

2Pos010 **MARTINI 粗視化シミュレーションにおけるタンパク質の多段階構造変化手法の開発**
Development of a method enabling multiple-step conformational change along a path in MARTINI coarse-grained simulations
Kaita Fujihara, Tatsuki Negami, Tohru Terada, Kentaro Shimizu (*Dept. of Biotech., Grad Sch. of Agri. Life Sci., Univ. of Tokyo*)

2Pos011 **構造に分布を持つ二重スピンラベルタンパクの電子スピン共鳴の緩和および線形の解析**
Relaxation and Lineshape Analysis for Electron Paramagnetic Resonance of Doubly Spin-labeled Protein with Structural Distribution
Yasunori Ohba[1], Munehito Arai[2], Jun Abe[3], Tetsuya Itabashi[1], Toshikazu Nakamura[3], Satoshi Takahashi[1], Seigo Yamauchi[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Art and Sci, Univ. Tokyo,*, [3]*IMS*)

2Pos012 **Structural insight into the interaction between Death-associated protein kinase 1 and natural flavonoids**
Yuto Kosaka, Mineyuki Mizuguchi, Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

2Pos013 **ウシ心筋チトクロム酸化酵素 O 型中間体の X 線結晶構造解析**
The X-ray structural analysis of O intermediate of bovine heart cytochrome *c* oxidase
Yuki Eto[1], Atsuhiro Shimada[1], Fumiyoshi Hara[2], Eiki Yamashita[2], Kyoko Shinzawa-Itoh[1], Tomitake Tsukihara[1,2], Shinya Yoshikawa[1] ([1]*Grad. Sch. Life Sci. Univ. Hyogo.*, [2]*Inst, Protein Res, Osaka Univ*)

2Pos014 **Molecular dynamics study of structural fluctuation in calmodulin**
Hiromitsu Shimoyama, Mayuko Takeda-Shitaka (*Kitasato University, Faculty of Pharmacy*)

2Pos015 **タンパク質立体構造予測のための新規な疎水性指標**
A novel measure for evaluating the satisfication of the hydrophobicity for protein structure prediction
Yota Masuyama, Koudai Takagi, George Chikenji (*Grad. Sch. Eng., Nagoya Univ.*)

2Pos016 **アミロイド線維凝集に関する分子動力学計算を用いた静電相互作用の研究**
Study of electrostatics interaction of amyloid fibrils using molecular dynamics calculation
Takuya Gouda[1], Yuko Okamoto[1,2,3,4] ([1]*Grad. Sch. Phys., Univ. Nagoya*, [2]*Struc.Biol.Res.Center, Grad.Sch.Sci., Nagoya Univ*, [3]*Center Comput. Sci., Grad. Sch. Eng.,Nagoya Univ*, [4]*Info. Tech. Center, Nagoya Univ*)

2Pos017 **α ヘリックスが β-ストランドペア形成に及ぼす影響**
The effect of α-helices on β-strand pairing propensity
Hiromi Suzuki (*Sch. Agri., Meiji Univ.*)

蛋白質：構造･機能 / Protein: Structure & Function

2Pos018 **分子動力学シミュレーションを用いた Gads の SLP-76 認識機構の解析**
Analysis of the SLP-76 ligand recognition mechanism of Gads by molecular dynamics simulation
Yoshiyuki Uemura, Kazuyoshi Ueda, Motoyasu Ozawa (*Yokohama National University*)

2Pos019 **Subunits kinetics in alpha-crystallin as studied by small angle neutron scattering**
Rintaro Inoue[1], Takumi Takata[1], Norihiko Fujii[2], Nobuhiro Sato[1], Yojiro Oba[1], Shinichi Takata[3], Noriko Fujii[1], Masaaki Sugiyama[1] ([1]*Research Reactor Institute,Kyoto University*, [2]*Teikyo Univ, Radioisotope Res Ctr,*, [3]*Japan Atom Energy Agency*)

2Pos020 **単粒子解析法を用いて 26S プロテアソームの立体構造解析**
Structural analysis of the 26S proteasome by cryo-electron microscopy and Single-Particle Analysis
Zhuo Wang[1], Yasuo Okuma[1], Daiske Kasuya[2], Kaoru Mitsuoka[3], Yasushi Saeki[4], Takuo Yasunaga[1] ([1]*Department of Bioscience and Bioinformatics, Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [2]*Biomedicinal Information Research Center, Japan Biological Information Consortium (JBIC)*, [3]*Osaka University*, [4]*Laboratory of Protein Metabolism, Tokyo Metropolitan Institute of Medical Science*)

2Pos021 **TRAF 結合分子の結晶構造と X 線小角散乱解析**
Crystal structure and small-angle X-ray scattering analysis of TRAF binding protein
Teruya Nakamura[1], Chie Hashikawa[1], Yuya Yokote[1], Mami Chirifu[1], Yuu Taguchi[2], Jin Gohda[2], Taishin Akiyama[2], Kentaro Semba[3], Yoshinari Okamoto[1], Shinji Ikemizu[1], Masami Otsuka[1], Jun-ichiro Inoue[2], Yuriko Yamagata[1] ([1]*Grad. Sch. Pharmaceut. Sci., Kumamoto Univ.*, [2]*Inst. Medical Sci., Univ. Tokyo*, [3]*Dept. Life Sci. and Medical Bio-sci., Waseda Univ.*)

2Pos022 **緩和モード解析による転移温度付近の 10 残基のシニョリンのダイナミクス**
Dynamics of 10-Residue Peptide, Chignolin near a Transition Temperature using Relaxation Mode Analysis
Ayori Mitsutake[1,2], Hiroshi Takano[1] ([1]*Dep. of Phys., Keio Univ.*, [2]*JST, Presto*)

2Pos023 **大規模構造変化を許容するヘモグロビン結晶：結晶中タンパク質の動きの観測を目指して**
Hemoglobin crystals that allow large-scale conformational changes: toward the observation of protein motions in crystals
Naoya Shibayama (*Div. of Biophysics, Jichi Medical Univ.*)

2Pos024 **Circadian timing governed by cyanobacterial KaiC ATPase**
Atsushi Mukaiyama[1,2], Jun Abe[1], Takuya Hiyama[1], Seyoung Son[3], Toshifumi Mori[2,4], Shinji Saito[2,4], Masato Osako[3], Julie Wolanin[1], Eiki Yamashita[5], Takao Kondo[3], Shuji Akiyama[1,2] ([1]*CIMoS, IMS*, [2]*Grad. Univ. for Adv. Studies, SOKENDAI*, [3]*Nagoya Univ.*, [4]*Dep. of Theor. and Comp. Molecular Science, IMS*, [5]*IPR, Osaka Univ.*)

2Pos025 **サチライシンにおける局所的な柔軟性増大のアロステリック効果**
Allosteric effect of locally-enhanced flexibility in subtilisin
Takato Sato, Kosei Maetani, Dan Parkin, Jun Ohnuki, Koji Umezawa, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

2Pos026 **シナプス小胞膜を模倣した膜上での α シヌクレイン線維形成メカニズム**
Study on the mechanism of amyloidogenesis of α-synuclein on presynaptic membrane mimetics
Mayu Terakawa S.[1], Yuxi Lin[1], Tatsuya Ikenoue[1], Naoya Fukui[2], Yasushi Kawata[2], Young-Ho Li[1], Yuji Goto[1] ([1]*IPR, Osaka Univ.*, [2]*Dept. of Chem. and Biotech., Grad. Sch. of Eng., Tottori Univ.*)

2Pos027 **アナモックス菌のラダラン脂質生合成にかかわると推定されるラジカル SAM 酵素の発現と精製**
Expression and purification of a radical SAM enzyme, which is presumably involved in ladderane lipid biosynthesis in anammox bacteria
Jumpei Shimada[1], Nozomi Shinde[1], Tomoya Hino[1], Miki Fukuma[1], Ken Takai[2], **Shingo Nagano**[1] ([1]*Grad. Schl. Eng., Tottori Univ.*, [2]*JAMSTEC*)

2Pos028 **揺らぎと水和効果に着目した ABC トランスポーターの NBD 二量体化過程の統計熱力学的解析**
Statistical thermodynamic analysis of dimerization of nucleotide binding domains in an ABC transporter: fluctuation and hydration effects
Honami Sakaizawa, Hiroshi C. Watanabe, **Tadaomi Furuta**, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech*)

2Pos029 **区分同位体標識を用いた β_2-アドレナリン受容体のリン酸化の NMR 解析**
NMR analyses of the phosphorylation of β_2-adrenergic receptor using segmental isotopic labeling
Yutaro Shiraishi[1], Yutaka Kofuku[1], Takumi Ueda[1,2], Mei Natsume[1], Hideo Iwai[3], Ichio Shimada[1] ([1]*Grad.Sch.Pharm.Sci.,Univ.Tokyo*, [2]*JST,PRESTO*, [3]*Inst.Biotech.,Univ.Helsinki*)

2Pos030 **ホタルの発光に関与する酵素ルシフェラーゼの荷電性アミノ酸残基に対する滴定曲線の導出**
Computational Analysis of Titration Curves of Some Amino Acid Residues in Proximity to Catalytic Center of Firefly Luciferase
Naohisa Wada[1], Keiici Horie[1], Sho Takamatsu[1], Itsuki Kaji[1], Hironori Sakai[2] ([1]*Fac. of Food Life Sciences, Toyo Univ.*, [2]*Insti. of Fluid Science, Tohoku Univ.*)

2Pos031 **阻害剤 BOF を結合したバクテリア XOR のリガンド結合ポケット入口に存在する一つのループの揺らぎについて**
On the motion of a loop located in the entrance to the ligand-binding pocket of bacterial XOR with the inhibitor BOF
Hiroto Kikuchi[1], Hiroshi Fujisaki[1], Tadaomi Furuta[2], Ken Okamoto[3], Takeshi Nishino[4] ([1]*Dept. of Phys., Nippon Med. Sch.*, [2]*Grad. Sch. of Biosci. & Biotech., Tokyo Inst. of Tech.*, [3]*Dept. of Biochem., Nippon Med. Sch.*, [4]*Grad. Sch. of Agri. & Life Sci., Univ. Tokyo*)

2Pos032 **等温滴定型熱量測定によるヒトヘモグロビンーIHP 間の相互作用の研究**
Interactions between inositol hexakisphosphate and hemoglobin studied by isothermal titration calorimetry and oxygen binding measurements
Shunsuke Sakurai[1], Daiki Sawada[1], Tsuyoshi Egawa[2], Takashi Yonetani[3], Antonio Tsuneshige[1] ([1]*Dept. of Frontier Bioscience, Hosei University, Tokyo, Japan*, [2]*Dept. of Biochemistry, Albert Einstein School of Medicine, New York, USA*, [3]*Dept. of Biochemistry and Biophysics, Perelman School of Medicine, University of Pennsylvania, Philadelphia, USA*)

2Pos033 **3D-RISM 理論に基づく Pim1-リガンド系における結合自由エネルギーの予測**
Estimation of binding free energy based on the 3D-RISM theory for the Pim1-ligand system
Takeshi Hasegawa[1], Masatake Sugita[1], Takeshi Kikuchi[1], Fumio Hirata[2] ([1]*Dept. of Bioinf., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Research Org. of Sci. and Tech., Ritsumeikan Univ.*)

2Pos034 **粗視化モデルを用いた T7 RNA ポリメラーゼの転写動態に関する考察**
Study of transcriptional dynamics of T7 RNA polymerase using coarse-grained model
Kizuku Yamana, Hiraku Nishimori, Akinori Awazu (*Dept. of Math. and Life Sciences, Hiroshima Univ.*)

蛋白質：特性 / Protein: Property

2Pos035 **蛋白質熱安定性に及ぼす共溶媒の添加効果**
Effects of cosolvent addition on the thermal stability of a protein
Shota Murakami[1], Masahiro Kinoshita[2] ([1]*Grad. Sch. Energ. Sci., Kyoto Univ.*, [2]*Inst. Adv. Energ., Kyoto Univ.*)

2Pos036 **独立成分分析 tICA を用いたヒストンテールの遅い運動の解析**
Slow dynamics of histone tails in molecular dynamics simulation revealed by time-structure based independent component analysis
Sotaro Fuchigami (*Grad. Sch. of Medical Life Science, Yokohama City Univ.*)

2Pos037 **接着結合における張力伝達分子としての β-カテニン**
β-catenin as a mechano-transmitter molecule at adherens junctions
Koichiro Maki[1,2], Sung-Woong Han[1,2], Taiji Adachi[1,2] ([1]*Inst. for Front. Med. Sci., Kyoto Univ.*, [2]*Grad. Sch. of Eng., Kyoto Univ.*)

2Pos038 **Spectroscopic and calorimetric analysis for conformational stability of c-Myb DNA-binding domain under different pH conditions**
Satomi Inaba[1], Yuji O. Kamatari[2], Hiroshi Sekiguchi[3], Harumi Fukada[4], Masayuki Oda[1] ([1]*Grad. Sch. Life Environ. Sci., Kyoto Pref. Univ.*, [2]*Life Sci. Res. Center, Gifu Univ.*, [3]*JASRI/SPring-8*, [4]*Grad. Sch. Life Environ. Sci., Osaka Pref. Univ*)

2Pos039 **高速時間分割蛍光異方性によるタンパク質間相互作用の解析**
Protein-protein interaction revealed by time-resolved fluorescence anisotropy
Akane Kato[1], Etsuko Nishimoto[2] ([1]*Grad. Sch. Bioresour. Bioenviron. Sci., Univ. Kyushu*, [2]*Fac. Agr., Univ. Kyushu*)

2Pos040 **高圧小角散乱法により検出されたニトリラーゼ会合体の高圧中間体解析**
Analysis on pressure intermediate of Nitrilase oligomer detected by high-pressure small-angle scattering
Tetsuro Fujisawa[1,2,3], Ryo Ishiguro[1,2] ([1]*Dep. Eng., Gifu Univ.*, [2]*Harima Inst., Riken*, [3]*NSSR, Nagoya Univ.*)

2Pos041 **フェリチンアセンブリ反応における静電相互作用の重要性**
Importance of electrostatic interactions during ferritin assembly reaction
Daisuke Sato, Atsushi Kurobe, Satsuki Takebe, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. Bioinfo., Grad. Sch. Eng., Soka Univ.*)

2Pos042 **アクチンフィラメントの圧電特性 II**
Piezoelectric property of an actin filament II
Jun Ohnuki[1], Takato Sato[1], Taro Q.P. Uyeda[2], Mitsunori Takano[1] ([1]*Dept. of Pure & Appl. Phys., Waseda Univ.*, [2]*Biomedical Res. Inst., AIST*)

2Pos043 **天然変性領域である p53C 末端ドメインの構造と物性にアセチル化が及ぼす影響**
Effect of acetylation on structure and physicochemical properties of the intrinsically disordered C-terminal domain of p53
Shinji Iida[1,2], Hironobu Hojo[1], Haruki Nakamura[1], Junichi Higo[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*)

2Pos044 **天然タンパク質の分子密度に関する統計解析**
Statistical analysis on the molecular density of native proteins
Hidenobu Kawai, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

2Pos045 **ALS を引き起こす SOD1 の線維形成に伴う物性変化**
Physical property change associated with fibril formation of ALS-causative SOD1
Noriko Fujiwara[1], Michiru Wagatsuma[2], Daisaku Yoshihara[1], Yoshiaki Furukawa[3], Haruhiko Sakiyama[1], Hironobu Eguchi[1], Keiichiro Suzuki[1] ([1]*Dep. of Biochem., Hyogo coll. of Med.*, [2]*Biosensor, Res. and Dev., ULVAC*, [3]*Dept. of Chem., Keio Univ.*)

2Pos046 **構造エレメントの決定因子としての分子内相互作用**
Intramolecular interaction as a determination factor for structure elements of proteins
Yuji Konno, Takeshi Inagaki, Hironari Kamikubo, Mikio Kataoka (*Graduate School of Materials Science, Nara Institute of Science and Technology*)

蛋白質：機能 / Protein: Function

2Pos047 **示差走査型蛍光定量法を用いたアルドケト還元酵素阻害剤の選択性の評価**
Evaluation of selectivity of aldo-keto reductase inhibitors using differential scanning fluorimetry
Yuji O. Kamatari[1], Aurangazeb Kabir[2], Satoshi Endo[3], Kazuo Kuwata[2] ([1]*Life Sci. Res. Center, Gifu Univ.*, [2]*United Grad. Sch. Drug Discov. Med. Info. Sci., Gifu Univ.*, [3]*Lab. Biochem., Gifu Pharm. Univ.*)

2Pos048 **GlcNAc 含有高分子を用いたビメンチンの新たな機能の解明**
Elucidation of novel functions of vimentin with the well-defined GlcNAc-bearing polymers
Sadanori Yamasaki[1], Horihiko Ise[2], Tadashi Nakamura[1], Yoshiko Miura[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Univ. Kyushu*, [2]*IMCE., Univ. Kyushu*)

2Pos049 **Protective effect of model peptides for group-3 LEA proteins on enzymes during desiccation**
Takao Furuki, Minoru Sakurai (*Center for Biological Resources and Informatics, Tokyo Institute of Technology*)

2Pos050 **NMR と分子動力学シミュレーションによるラン藻由来アルカン合成酵素の構造ダイナミクス解析**
Structural dynamics of a cyanobacterial alkane-producing enzyme, aldehyde deformylating oxygenase, studied by NMR and MD simulations
Yuma Suematsu, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

2Pos051 **四量体型サルコシン酸化酵素の分子動力学シミュレーション :生成物の輸送経路の解明**
Molecular dynamics simulation of heterotetrameric sarcosine oxidase: analysis of channeling of product
Daisuke Nakajima[1], Go Watanabe[2], Haruo Suzuki[2], Shigetaka Yoneda[2] ([1]*Grad. Sch. Sci., Kitasato Univ.*, [2]*Sch. Sci., Kitasato Univ.*)

蛋白質：計測･解析 / Protein: Measurement & Analysis

2Pos052 **生体高分子中性子結晶構造解析における水素の高感度検出のためのラジカル分子のタンパク質単結晶への導入**
An introduction of radical molecules into a protein single crystal for more sensitive detection of hydrogen in neutron crystallography
Naoya Komatsuzaki[1], Takahiro Iwata[2], Yoshiyuki Miyachi[2], Toshiyuki Chatake[3], Katsuhiro Kusaka[4], Nobuo Niimura[4], Ichiro Tanaka[4,5] ([1]*Grad. Sch. of Sci. and Eng., Ibaraki Univ.*, [2]*Faculty of Sci., Yamagata Univ.*, [3]*RRI, Kyoto Univ*, [4]*Frontier Res. Center*, [5]*Coll. of Eng., Ibaraki Univ.*)

2Pos053 **Raf の生細胞内構造分布を明らかにする 1 分子 FRET 計測**
Single-molecule FRET measurement to investigate structural distribution of Raf in living cells
Kenji Okamoto[1], Kayo Hibino[2], Yasushi Sako[1] ([1]*RIKEN*, [2]*NIG*)

2Pos054 **On the convergence of binding free-energy landscape calculation by improved virtual-system coupled adaptive umbrella sampling**
Bhaskar Dasgupta, Junichi Higo, Haruki Nakamura (*Institute for Protein Research, Osaka University*)

2Pos055 **X 線 1 分子追跡法によるミオシン分子構造揺らぎの高精度リアルタイム計測**
Molecular fluctuation analysis of myosin by DXT measurement
Keigo Ikezaki[1], Hiroshi Sekiguchi[2], Naoto Yagi[2], Toshio Yanagida[3], Yuji C. Sasaki[1,2] ([1]*Tokyo Univ.*, [2]*Spring-8/JASRI*, [3]*RIKEN/QBiC*)

2Pos056 **フェリチンからの鉄イオン遊離に与える交流磁場の影響**
Iron exit from ferritin under alternating magnetic fields
Yuta Yamada, Tsuyoshi Hondou, **Hidetake Miyata** (*Facuty of Science, Tohoku University*)

2Pos057 **1 分子光子計数データのデータ同化解析**
Sequential data assimilation for single-molecule FRET photon-counting data
Yasuhiro Matsunaga[1], Yuji Sugita[1,2,3,4] ([1]*RIKEN AICS*, [2]*RIKEN*, [3]*RIKEN iTHES*, [4]*RIKEN QBiC*)

蛋白質：工学 / Protein: Engineering

2Pos058 **がん関連サイトカインに結合するラクダ科単一ドメイン抗体の試験管内淘汰と、その簡便・迅速なプルダウン法**
***In vitro* selection and a simple and rapid pull-down assay of Camelidae single-domain antibodies that bind to cancer cytokine**
Hidenao Arai, Mana Mizugai, Koji Matsuoka, Naoto Nemoto (*Grad. Sch. Sci. Eng., Saitama Univ.*)

2Pos059 **α ヘリカルタンパク質の設計**
Design of diverse all-α protein strucrures based on αα-rule
Kouya Sakuma[1,2], Rie Koga[1], Nobuyasu Koga[1,2,3] ([1]*CIMoS, IMS.*, [2]*SOUKENDAI*, [3]*JST, PRESTO*)

2Pos060 **An attempt to improve the method for directed evolution by in vitro compartmentalization**
Kentaro Tahara[1], Ryo Iizuka[1], Ayaka Iguchi[2], Hyun Yoon Dong[2], Tetsushi Sekiguchi[2], Shuichi Shoji[2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Adv. Sci. and Eng., Waseda Univ.*)

2Pos061 **Incorporation of a photoisomerizable non-natural amino acid into proteins for photo-control of protein functions**
Rumi Shiba, Takayoshi Watanabe, Takahiro Hohsaka (*School of Materials Science, Japan Advanced Institute of Science and Technology*)

ヘム蛋白 / Heme proteins

2Pos062 **癌抑制タンパク質 101F6 のアスコルビン酸からの電子受容機構**
Electron transfer mechanism from ascorbate to human tumor suppressor 101F6 protein
Fusako Takeuchi[3], Takako Yamazoe[1], Hiroaki Okano[1], C. Mariam Recuenco[1,2], Takahiro Kozawa[4], Kazuo Kobayashi[4], **Motonari Tsubaki**[1] ([1]*Chemistry, Grad. Sch. Sci., Kobe Univ.*, [2]*Univ. Philip., Los Banos*, [3]*IPHE, Kobe Univ.*, [4]*ISIR, Osaka Univ.*)

2Pos063 **ハーフメトヘモグロビン M を用いたヒト成人ヘモグロビンの酸素親和性制御に関する α 鎖と β 鎖の Fe-His 結合の役割の違いに関する研究**
Study on different roles of Fe-His bond between α and β chains for oxygen affinity regulation of human hemoglobin by using half-met Hb Ms
Shigenori Nagatomo[1], Kazuya Saito[1], Masako Nagai[2], Takashi Ogura[3], Teizo Kitagawa[3] ([1]*Dept. Chem., Univ. Tsukuba*, [2]*Res. Center Micro-Nano Tech., Hosei Univ.*, [3]*Grad. Sch.Life Sci., Univ. Hyogo*)

2Pos064 **酸素センサー酵素 YddV のヘム結合グロビンドメインの結晶構造解析**
Crystal structure of isolated heme-bound globin domain of a heme-based oxygen-sensor enzyme, YddV, from *Escherichia coli*
Jotaro Igarashi, Toru Kikuchi, Ariki Matsuoka (*Dept. Nat. Sci., Biol., Sch. of Med., Fukushima Med. Univ.*)

2Pos065 **チトクロム酸化酵素に対するチトクロム c の 2 つの結合部位の X 線構造解析**
X-ray structural analysis of two binding sites of cytochrome *c* to cytochrome *c* oxidase
Satoru Shimada[1,2], Shimpei Aoe[1], Kyoko Shinzawa-Itoh[1], Junpei Baba[1], Atsuhiro Shimada[1], Eiki Yamashita[3], Shinya Yoshikawa[1], Tomitake Tsukihara[1,2,3] ([1]*Picobiology Inst., Grad. Sch. Life Sci., Univ. Hyogo*, [2]*CREST, JST*, [3]*Inst. Protein Res., Osaka Univ.*)

膜蛋白質 / Membrane proteins

2Pos066 **三量体ハロロドプシンにおけるカロテノイド結合能の特異性**
Specificity of carotenoid binding to halorhodopsin trimer
Yasuyuki Miyazaki[1], Takashi Kikukawa[2], Makoto Demura[2], Noritaka Kato[1], Takanori Sasaki[1] ([1]*Sch. Sci. and Tech., Meiji Univ.*, [2]*Adv. Life Sci., Hokkaido Univ.*)

2Pos067 **脂質平面膜法を用いた TRPM3 チャネルの機能解析**
Single channel analysis of the TRPM3 channels in planar lipid bilayers
Kunitoshi Uchida[1,2,3], Lusine Demirkhanyan[2], Eleonora Zakharian[2], Makoto Tominaga[1,3] ([1]*Div of Cell Signal, OIIB (NIPS)*, [2]*Dept of Cancer Biol and Pharmacol, Univ of Illinois Coll of Med.*, [3]*Dept of Physiol Sci, SOKENDAI*)

2Pos068 **分子シミュレーションによる阻害剤が結合した ADP/ATP carrier の内向き開構造**
Structure of the inhibitor-bound inward-facing conformation of ADP/ATP carrier: A simulatioin study
Koichi Tamura[1], M. Harunur Rashid[1,2], Shigehiko Hayashi[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*RMIT Univ., Melbourne, Australia*)

2Pos069 **ケイジド化合物導入による低分子量 G タンパク質 H-Ras の多量体形成の光制御**
Photo-control of small G-protein H-Ras multimerization using caged compounds
Seigo Iwata[1], Takashi Hashimoto[2], Nobuhisa Umeki[3], Kazunori Kondo[2] ([1]*Grad. Sch. Eng., Univ. SOKA*, [2]*Fac. Eng., Univ. SOKA*, [3]*Wasko Inst., Riken*)

2Pos070 **細菌外膜シトクロムにおける代謝共役したフレキシブルな構造と機能**
Flexible Strucutral Alteration Associated with Respriatory Electron Flow in Microbial Outer Membrane c-type Cytochrome Complex
Akihiro Okamoto, Yoshihide Tokunou, Chinotaikul Punthira, Kazuhito Hashimoto (*Grad. Sch. Eng., Univ. Tokyo*)

2Pos071 **結晶化を目指したカイコガ性フェロモン生合成活性化神経ペプチド受容体（PBANR）のアンタゴニストの探索**
Ligand screening of silkmoth pheromone biosynthesis-activating neuropeptide receptor for crystallization of the ligand-receptor complex
Yukie Katayama[1], Hidekazu Katayama[2], Takeshi Kawai[1], Tatsuya Suzuki[1], Tatsuki Ebisawa[1], Ryo Natsume[3], Yu-Hua Lo[4], Toshiya Senda[4], Toshihiro Nagamine[5], Masaaki Kurihara[5], Jae Min Lee[5], Joe J. Hull[6], Shogo Matsumoto[5], Hiromichi Nagasawa[1], Koji Nagata[1], Masaru Tanokura[1] ([1]*UTokyo*, [2]*Tokai Univ.*, [3]*TDU*, [4]*KEK-PF*, [5]*RIKEN*, [6]*USDA-ARS*)

2Pos072 **チャネル電流計測を用いた脂質二重膜中における L-, D-ボンビニンの抗菌活性の解析**
Antimicrobial activity analysis of L- and D-Bombinin in lipid bilayers using channel current recording
Yusuke Sekiya[1], Hirokazu Watanabe[1], Yuki Kitahashi[2], Izuru Kawamura[2], Ryuji Kawano[1] ([1]*Tokyo Univ Agr Tech*, [2]*Grad. Sch. Eng, Yokohama Nattl Univ.*)

2Pos073 **Active supercomplex purified from bovine heart reveals the functional unit of the mitochondrial respiratory chain**
Kyoko Shinzawa-Itoh[1], Harunobu Shimomura[1], Sachiko Yanagisawa[1], Satoru Shimada[1,2], Ryoko Takahashi[1], Shigefumi Uene[1], Takashi Ogura[1], Shinya Yoshikawa[1], Tomitake Tsukihara[1,2] ([1]*Grad. Sch. Life Sci., Univ. Hyogo*, [2]*CREST, JST*)

2Pos074 **上皮成長因子受容体（EGFR）の膜貫通および膜近傍ドメインの構造ダイナミクス解析**
Conformational dynamics of transmembrane and juxtamembrane domains of epidermal growth factor receptor (EGFR)
Ryo Maeda[1], Takeshi Sato[2], Kenji Okamoto[1], Takehiko Inaba[3], Yasushi Sako[1] ([1]*Cellular Informatics Lab., RIKEN*, [2]*Inst. for Protein Research, Osaka Univ.*, [3]*Lipid Biology Lab., RIKEN*)

核酸結合蛋白質 / Nucleic acid binding proteins

2Pos075 **Characterization of RuvA-RuvB-Holliday junction DNA complex formation using Zero Mode Waveguides**
Yong-Woon Han[1], Takuma Iwasa[1,2], Ryo Hiramatsu[3], Hiroaki Yokota[1,4], Kimiko Nakao[1], Ryuji Yokokawa[5,6], Teruo Ono[3], Yoshie Harada[1,2] ([1]*iCeMS, Kyoto University*, [2]*Grad. Sch. Biostudies, Kyoto Univ.*, [3]*Inst. for Chem. Res., Kyoto Univ.*, [4]*Grad. Sch. for the Creation of New Photonics Ind.*, [5]*Grad. Sch. Tech., Kyoto Univ.*, [6]*PREST*)

2Pos076 **The stability of the translation arrest of SecM internal deletion mutants**
Yuanfang Guo, Zhuohao Yang, Ryo Iizuka, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

2Pos077 **高活性 TALEN の高活性化機構の解析**
In vitro analysis of super-active TALEN
Yoko Terahara[1], Kazuho Ikeda[1], Naoyuki Miyasita[2], Yasushi Okada[1] ([1]*QBic, Riken*, [2]*CDB, Riken*)

核酸：構造･特性 / Nucleic acid: Structure & Property

2Pos078 **弾性ネットワークモデルによる DNA の配列・構造・運動と機能の関係の考察**
Analysis of the relationship among sequences, structures, motions and the functions of DNA by elastic network models
Shuhei Isami[1], Sayuri Tatemoto[1], Atsushi Ikegaya[1], Hiraku Nishimori[1,2], Naoaki Sakamoto[1,2], Akinori Awazu[1,2] ([1]*Dept. Math. and Life Sciences, Hiroshima Univ.*, [2]*Research Center for Mathematics on Chromatin Live Dynamics*)

2Pos079 **DNA 二重鎖をほどく１分子計測とシミュレーションによる水素結合の構造へのエントロピー寄与**
Entropic effects of hydrogen bonds to the double-stranded DNA structure revealed by single-molecule mechanical unzipping
Akihiro Fukagawa[1], Michio Hiroshima[2], **Makio Tokunaga**[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*RIKEN*)

2Pos080 **磁場によるプラスミド DNA への影響**
The effect of magnetic fields on the plasmid DNA
Masahiko Hasegawa, Tsuyoshi Hondou, Hidetake Miyata (*Department of Physics, Tohoku University Graduate School*)

2Pos081 **DNA 高次構造転移における３価陽イオンと２価陽イオンの競合効果**
Mutual inhibition between 2+ and 3+ cations to induce folding transition of DNA
Chika Tongu[1], Takahiro Kenmotsu[1], Yuko Yoshikawa[2], Kenichi Yoshikawa[1] ([1]*Doshisha Univ.*, [2]*Ritsumeikan Univ.*)

2Pos082 **熱ショックタンパク質の発現調節をする温度感知 RNA の分子動力学シミュレーション**
Molecular dynamics simulations of a temperature-sensitive RNA that regulates the gene expression of heat-shock proteins
Yoshiharu Mori[1], Hisashi Okumura[1,2] ([1]*IMS*, [2]*SOKENDAI*)

核酸：相互作用･複合体形成 / Nucleic acid: Interaction & Complex formation

2Pos083 **DNA および酵素のハイドロゲルカプセル内封入による単純なセルオートマトンの化学実装に向けて**
Toward chemically implementing simple cellular automaton by encapsulating DNA and Enzyme in hydrogel capsule
Ibuki Kawamata[1], Satoru Yoshizawa[1], Fumi Takabatake[1], Ken Sugawara[2], Satoshi Murata[1] ([1]*Tohoku University*, [2]*Tohoku Gakuin University*)

2Pos084 **DNA のよじれ合いと巻き付きにおけるカイラリティの選択性**
Chiral Selection in Braiding and Wrapping of Double-Stranded DNA
Sosuke Sano[1], Tomohiro Yanao[1], Kenichi Yoshikawa[2] ([1]*Grad. Sch. Sci. & Eng., Waseda Univ.*, [2]*Grad. Sch. Life & Med. Sci., Doshisha Univ.*)

電子状態 / Electronic state

2Pos085 **オーダーN法第一原理分子動力学計算のデモンストレーション**
Demonstration of order-N first-principles density functional theory-molecular dynamics calculations
Takao Otsuka[1], Michiaki Arita[2], Makoto Taiji[1], David R. Bowler[3], Tsuyoshi Miyazaki[4] ([1]*RIKEN QBiC*, [2]*Grad. Sch. Sci. and Tech., Tokyo Univ. of Sci.*, [3]*UCL*, [4]*NIMS*)

水･水和･電解質 / Water & Hydration & Electrolyte

2Pos086 **一般化ボルンモデルにおける塩橋の安定性**
Salt bridge stability in the generalised Born model
Dan Parkin, Yukinobu Mizuhara, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

2Pos087 **剛体球参照系を用いた密度汎関数理論に基づく新しい溶媒和自由エネルギー表式**
A Solvation-Free-Energy Functional: A Reference-Modified Density Functional Formulation
Yutaka Maruyama[1], Tomonari Sumi[2], Ayori Mitsutake[3] ([1]*AICS, RIKEN*, [2]*Dep. Chem., Okayama Univ.*, [3]*Dep. Phys., Keio Univ.*)

2Pos088 **MM/3D-RISM 法を用いた 2-Hydroxypropyl-β-Cyclodextrin の抱接現象における結合自由エネルギーの解析**
Analyzing the binding free energy for the inclusion process of 2-Hydroxypropyl-β-Cyclodextrin by means of the MM/3D-RISM method
Masatake Sugita[1], Fumio Hirata[2] ([1]*Dept. of Bioinfo., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Research Org. of Sci. and Tech., Ritsumeikan Univ.*)

発生･分化 / Development & Differentiation

2Pos089 **機械的な力による上皮形態形成制御**
Mechanical control of epithelial morphogenesis
Kaoru Sugimura[1,2], Keisuke Ikawa[1], Shuji Ishihara[3] ([1]*iCeMS, Kyoto Univ.*, [2]*JST PRESTO*, [3]*Fac. of Sci. and Eng., Meiji Univ.*)

2Pos090 **血管新生における内皮細胞動態ライブイメージング**
Live cell imaging of vascular endothelial cell dynamics in angiogenesis
Naoko Takubo[1], Kazuaki Naemura[1], Ryo Yoshida[2], Terumasa Tokunaga[3], Osamu Hirose[4], Yasunobu Uchijima[1], Yukiko Kurihara[1], Hiroki Kurihara[1] ([1]*Graduate School of Medicine, The University of Tokyo*, [2]*The Institute of Statistical Mathematics, Research Organization of Information and Systems*, [3]*Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [4]*College of Science and Engineering, Kanazawa University*)

2Pos091 **胎生期脳組織に発現するメカノトランスダクション関連遺伝子の解析**
The functional analysis of the mechanotransduction-related gene expressed in the developing brain
Misato Iwashita[1], Noriyuki Kioka[3], Yoichi Kosodo[2] ([1]*CDB, RIKEN*, [2]*Korea Brain Research Institute*, [3]*Grad. Sch. Agri., Kyoto Univ.*)

筋肉 / Muscle

2Pos092 **水中におけるアクチン重合の統計熱力学について**
On the statistical thermodynamics of actin polymerization in aqueous solution
Tomohiko Hayashi[1], Hiraku Oshima[1], Makoto Suzuki[2], Masahiro Kinoshita[1] ([1]*Inst. Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Eng., Tohoku Univ.*)

2Pos093 **ESR 距離測定による心筋トロポニン I と T のリン酸化調節の 動的構造基盤**
Structural dynamics of cardiac troponin regulated by phosphorylation, as studied by distance measurement using spin-labeling dipolar EPR
Hiroaki Yamashita[1], Chenchao Zhao[1], Koichi Sakai[1], **Keisuke Ueda**[1], Tatsuhito Matsuo[2], Satoru Fujiwara[2], Shoji Ueki[3], Toshiaki Arata[1] ([1]*Dept. Biol., Sci. Grad. Sch., Osaka Univ.*, [2]*JAEA*, [3]*Tokushima-Bunri Univ.*)

2Pos094 **ミオシンフィラメント懸濁液の ADP 存在・非存在下でのプロトン NMR 緩和経過**
Spin-spin relaxation of 1H NMR signals from myosin filaments suspension with or without ADP
Tetsuo Ohno, Toshiko Yamazawa (*Department of Physiology The Jikei University School of Medicine*)

2Pos095 **OH 伸縮振動のラマン分光によるミオシン S1 の水和状態の測定**
Hydration study on myosin subfragment-1 (S1) by Raman OH-stretching spectroscopy
Yuki Ochiai[1], Hideyuki Ohsugi[1], George Mogami[1], Tetsuo Taniuchi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Univ. Tohoku*, [2]**FRIS., Univ. Tohoku*)

2Pos096 **光学顕微鏡および放射光 X 線回折実験による甲虫飛翔筋の構造的考察**
Structural insights in coleopteran flight muscles by optical microscopy and synchrotron X-ray diffraction measurements
Toshiki Shimomura[1], Hiroyuki Iwamoto[2], Hirotaka Sato[3], Madoka Suzuki[4,5], Shin'ichi Ishiwata[1,4,5] ([1]*Sch Adv Sci Eng, Waseda Univ*, [2]*SPring-8, JASRI*, [3]*Sch Mech Aerospace Eng, Nanyang Tech Univ (NTU), Singapore*, [4]*WASEDA Biosci Res Inst Singapore (WABIOS)*, [5]*Org Univ Res Initiatives, Waseda Univ*)

2Pos097 **Mg/Ca 結合 F-アクチンの多重水和状態**
Multi-hydration States of F-actin Bound with Mg/Ca Ions
Ryotaro Chishima[1], Asato Imao[1], George Mogami[1], Takahiro Watanabe[1], Tetsuichi Wazawa[2], Nobuyuki Morimoto[1], Makoto Suzuki[1,3] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Inst. Sci. Ind. Res., Osaka Univ.*, [3]*FRIS, Tohoku Univ.*)

分子モーター / Molecular motor

2Pos098 **In-plane 2D buckling of microtubule under compressive stress**
Arif Md. Rashedul Kabir[1], Daisuke Inoue[1], Kazuki Sada[1,2], **Akira Kakugo**[1,2] ([1]*Fac.of Sci., Hokkaido Univ.*, [2]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*)

2Pos099 **Emergence of ultra-large vortex pattern by collectively moving microtubules on application of external indentation stimuli**
Tamanna Ishrat Farhana[1], Daisuke Inoue[2], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

2Pos100 **ATP 存在下のミオシン S1 はアクチンフィラメントのらせんピッチを伸ばしコフィリン結合を阻害する：高速 AFM による直接観察**
Active myosin S1 induces longer helical pitches of actin filaments and inhibits actin binding of cofilin as demonstrated by high speed AFM
Kien Ngo[1], Noriyuki Kodera[2], Taro Uyeda[1] ([1]*Biomed. Res. Inst., AIST*, [2]*BioAFM FRC., Kanazawa U.*)

2Pos101 **バクテリア由来セルラーゼ *Cellulomonas fimi* Cel6B の逐次運動の 1 分子蛍光可視化解析**
Single-molecule fluorescence imaging analysis of processive movement of a bacterial cellulase *Cellulomonas fimi* Cel6B
Daiki Ishiwata[1,2,3], Akihiko Nakamura[1,2,3], Tomoyuki Tasaki[4], Ryota Iino[1,2,3] ([1]*Okazaki Inst. for Integrative Bio.*, [2]*Inst. for Molecular Science*, [3]*Sch., Phis. Sci., SOKENDAI*, [4]*Sch. Eng., Univ. Tokyo*)

2Pos102 **Individual kinesin immobilization on microfabricated nano-pillar arrays**
Taikopaul Kaneko[1], Fumie Oda[1], Takahide Kon[3], Ryuji Yokokawa[2] ([1]*Grad. Sch. Eng., Univ. Kyoto*, [2]*Grad. Sch. Eng., Assoc. Prof, Univ. Kyoto*, [3]*Grad. Sch. Eng., Dep. Biol. Sci., Prof, Univ. Osaka*)

2Pos103 **細菌Ⅲ型分泌機構の解明を目指したエフェクター輸送のリアルタイム評価系の構築**

Real-time evaluation of effector transport for clarifying the transport mechanism on type III secretion apparatus

Takashi Ohgita, Tsubasa Uekawa, Kyoko Momiyama, Naoki Hayashi, Naomasa Gotoh, Kentaro Kogure (*Kyoto Pharm. Univ.*)

2Pos104 **電場印加によるアクトミオシンの運動速度の可逆的変化**

A reversible change in the velocity of the motility of actin filaments on a heavy meromyosin-coated surface under an electric field

Kuniyuki Hatori[1], Taiki Abe[1], Reito Wada[2] ([1]*Grad. Sch. Sci. Eng., Yamagata Univ.*, [2]*Grad. Sch. Med., Yamagata Univ.*)

2Pos105 **新規フォトクロミック阻害剤を用いたキネシン Eg5 の光可逆的制御**

Photo-reversible control of mitotic kinesin Eg5 using a novel photochromic inhibitor

Ryoma Yamamoto, Kei Sadakane, Yuhki Tamura, Kentaro Saito, Shinsaku Maruta (*Soka University Graduate School of Engineering Division of Bioinformatics*)

2Pos106 **新規イネキネシン E11 に関する速度論的研究**

Kinetic study on the novel rice plant kinesin E11

Hironobu Taniguchi[1], Naoto Inomoto[2], Shinsaku Maruta[1,2] ([1]*Division Of Bioinformatics, Graduate School Of Engineering, Soka University*, [2]*Department Of Bioinformatics, Faculty Of Engineering, Soka University*)

2Pos107 **速度改変型キメラミオシン XI の発現により明らかになってきた原形質流動の機能と制御**

Function and regulation of cytoplasmic streaming on plant development

Motoki Tominaga[1], Kohji Ito[2], Takeshi Haraguchi[2], Etsuo Yokota[3], Akihiko Nakano[4,5] ([1]*Fac. Educ. Integrated Arts and Sci., Waseda Univ.*, [2]*Grad. Sch. Sci., Univ. Chiba*, [3]*Grad. Sch. Sci., Univ. Hyogo*, [4]*Grad. Sch. Sci., Univ. Tokyo*, [5]*RAP, RIKEN*)

2Pos108 ***Mycoplasma pneumoniae*・P1 adhesin の構造と機能**

Structure and function of P1 adhesin of *Mycoplasma pneumoniae*

Yu Matsumoto[1], Yoshito Kawakita[1], Tsuyoshi Kenri[2], Shigetaro Mori[2], Tasuku Hamaguchi[1], Miki Kinoshita[1], Akihiro Kawamoto[3], Takayuki Kato[3], Keiichi Namba[3], Makoto Miyata[1] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Infect. Dis. Inst*, [3]*Grad. Sch. Front. Biosciences., Univ. Osaka*)

2Pos109 **F1-ATPase の P-loop 変異体 TF1（βG158A）とリン酸解離の関係**

The relationship between F1-ATPase P-loop mutant TF1(βG158A) and Pi release

Hiroka Narita[1], Hitoshi Hoshina[1], Hikaru Yoshida[1], Ayumi Ito[1], Jotaro Ito[1,3], Yohei Nakayama[1], Shoichi Toyabe[2], Hiroshi Ueno[1,3], Eiro Muneyuki[1] ([1]*Dept. of Physics. Chuo Univ.*, [2]*Faculty of Physics. Tohoku Univ.*, [3]*School of Engineering, The University of Tokyo*)

2Pos110 **分裂期 kinesin-6 の運動特性に関する研究**

A study on motility of mitotic kinesin-6

Yohei Maruyama[1], Akihiko Sato[1], Tim Davis[2], Toshihisa Osaki[3], Shin Yamaguchi[1], Shoji Takeuchi[3], Masanori Mishima[2], Junichiro Yajima[1] ([1]*Dept. Life Sci., Grad. Sch. of Arts and Sci., Univ. of Tokyo*, [2]*CMCB at Warwick Med. Sch., Univ. of Warwick*, [3]*Inst. of Ind. Sci., Univ. of Tokyo*)

2Pos111 **ヒト細胞質ダイニン 1 分子のパワーストローク運動と反応機構**

The power stroke mechanism of human cytoplasmic dynein revealed by optical tweezers

Yoshimi Kinoshita[1], Taketoshi Kambara[2], Kaori Nishikawa[1], Motoshi Kaya[1], Hideo Higuchi[1] ([1]*Graduate School of Science, The University of Tokyo*, [2]*RIKEN QBiC*)

2Pos112 **DNA オリガミバネを用いた負荷存在下でのキネシンの運動の蛍光一分子観察**

Single-molecule fluorescent observations of kinesin-1 moving under a load from DNA origami spring

Kouhei Matsuzaki[1], Mitsuhiro Iwaki[2,3,4], Michio Tomishige[1] ([1]*Dept. Appl. Phys.,Grad. Sch.Eng., Univ. Tokyo*, [2]*QBiC, RIKEN*, [3]*Harvard Med. Sch.*, [4]*Grad. Sch. Frontier Biosci., Osaka Univ.*)

2Pos113 **長円形 PDMS チャンバー内における微小管の動的不安定性**

Dynamic instability of microtubules observed in oval PDMS microchambers

Yu Onodera[1], Tomohiro Shima[2,3], Yasushi Okada[2], Tomoko Masaike[1,4,5] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Science*, [2]*QBiC, RIKEN*, [3]*Dept. Biol. Sci., Grad. Sch. of Sci., Univ. of Tokyo*, [4]*Res. Inst. for Sci. and Tech.,Tokyo Univ. of Science*, [5]*PRESTO, JST*)

2Pos114 **高分解能ステップ計測で分かってきた細菌べん毛モーターのサーマルラチェット機構**

Thermal ratchet mechanism of the bacterial flagellar motor emerged by high-resolution nanophotometry

Shuichi Nakamura[1,2], Yusuke V. Morimoto[2,3], Nobunori Kami-ike[2], Yoshiyuki Sowa[4], Tohru Minamino[2], Keiichi Namba[2] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*QBiC, RIKEN*, [4]*Dept. Frontier Biosci., Hosei Univ.*)

2Pos115 **好アルカリ性 *Bacillus* 属細菌が持つ Na^+駆動型べん毛モーター固定子 MotPS の中性環境でのイオン透過機構の解明**

Analysis of the reduced motility mechanism of the Na^+-driven flagellar motor stator MotPS in alkaline *Bacillus* at low pH

Yuka Takahashi[1], Yukina Noguchi[2], Masahiro Ito[1,2] ([1]*Bio-Nano., Toyo Univ.*, [2]*Grad. Sch. Life Sciences, Toyo Univ.*)

2Pos116 **人工繊毛モーターの創製ー微小管の波打ち運動発現条件ー**

Development of self-propelled artificial cilia constructed from biomolecular motors

Ren Sasaki[1], Shoki Wada[1], Masaki Ito[1], Daisuke Inoue[1], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. Chem. Sci. Eng., Hokkaido Univ*, [2]*Fac. Sci., Hokkaido Univ*)

2Pos117 **超好熱菌 *Aquifex aeolicus* の固定子タンパク質 MotA のナノディスク再構成とその機能解析**

Functional reconstitution of stator protein MotA from *Aquifex aeolicus* into Nanodisc

Mizuki Gohara[1], Norihiro Takekawa[1], Naoya Terahara[2], Takayuki Kato[2], Keiichi Namba[2,3], Yasuhiro Onoue[1], Michio Homma[1] ([1]*Divi. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch., Frontier Biosci., Osaka Univ.*, [3]*Riken QBiC*)

2Pos118 **生細胞内における微小管へのキネシン結合速度定数の直接計測**

Direct measurement of the binding rate constant of kinesin to microtubules in living cells

Taketoshi Kambara, Yasushi Okada (*Riken, Quantitative Biology Center*)

2Pos119 **Measurment of the transition between docking and undocking of kinesin neck linkers**
Yuichi Kondo, Hideo Higuchi (*Grad. Sci. Phys., Univ. Tokyo*)

細胞生物学 / Cell biology

2Pos120 **FRAP による、成長円錐のラメリポディア領域におけるファシンの動態解析**
Dynamics of fascin analyezed by FRAP, in the lamellipodial region of the growth cone
Minami Tanaka[1,2], Ryoki Ishikawa[3], Kaoru Katoh[2] ([1]*Grad. Sch. Life & Env. Sci., Univ. Tsukuba*, [2]*Bio Mes Res. Inst., AIST*, [3]*Gunma Pref.Coll. Health Sci*)

2Pos121 **Dynamical Patterns and Physical Environments in Vibrio alginolyticus Swarm Plate**
Tzu-Jung Hsu (*National Central University*)

2Pos122 ***Spiroplasma eriocheiris* において観察されたキンクの伝搬によって生じるらせん型スクリューの向きと回転速度の変化**
Change in handedness and rotational speed of helical screw of *Spiroplasma eriocheiris* driven by kink propagation
Tatsuro Itou[1], Yoshiaki Kinosita[1], Daisuke Nakane[1], Hirofumi Wada[2], Wen Wang[3], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin Univ.*, [2]*Department of Physics, Ritsumeikan Univ.*, [3]*College of Life Science, Nanjing Normal Univ.*)

2Pos123 **クライオ電子線トモグラフィ法を用いたフィロポディア内アクチンフィラメント束化メカニズム解明**
Revealing the bundling mechanisms of actin filament in filopodia with cryo-electron tomography
Shinji Aramaki[1], Kouta Mayanagi[2], Kazuhiro Aoyama[3,4], Takuo Yasunaga[1] ([1]*Dept. of Bioscience and Bioinformatics, Kyushu Inst. of Tech.*, [2]*Medical Inst. of Bioregulation Kyushu Univ.*, [3]*FEI Company*, [4]*Research Center for Ultra-High Voltage EM, Osaka Univ.*)

2Pos124 **マウス網膜細胞光シグナル伝達の生体分子混み合いを考慮した数理モデル**
Rate equation models of phototransduction influenced by molecular crowding into membranous disks of mouse rod cell
Rei Takamoto[1], Hiraku Nishimori[1,2], Akinori Awazu[1,2] ([1]*Department of Mathematical and Life Science, Hiroshima University*, [2]*Research Center for Mathematics on Chromatin Live Dynamics, Hiroshima University*)

2Pos125 **ケラトサイトのかたち形成におけるストレスファイバーの役割**
Stress fibers contribute to the shape determination in fish keratocytes
Takako Nakata[1], Chika Okimura[1], Takahumi Mizuno[2], **Yoshiaki Iwadate**[1] ([1]*Fac. Sci., Yamaguchi Univ.*, [2]*Biomed. Res. Inst., AIST*)

2Pos126 **アクチンフィラメントとアクチン結合タンパク質間の協同的結合の経時的観察**
Longitudinal observation of cooperative binding between actin-binding proteins and actin filament
Rika Hirakawa[1], Atsuki Yoshino[1], Keitaro Shibata[2], Hiroaki Ueno[1], Taiga Imai[1], Taro Q.P. Uyeda[2], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Sustain. Environ. Eng., Muroran Inst.*, [2]*Biomedical Res. Inst., AIST.*)

2Pos127 **Photo-regulation of small G protein RhoA using photochromic molecules**
Masahiro Kuboyama[1], Kaori Masuhara[2], Shinsaku Maruta[2] ([1]*SOKA University Department of Bioinfomatics*, [2]*SOKA University Graduate School of Engeneering*)

2Pos128 **局在した細胞間接着が生み出す細胞運動パターン**
Localized intercellular adhesion inducing moving cell pattern
Katsuyoshi Matsushita (*CMC, Osaka Univ.*)

2Pos129 **Structural characterization of FliP, a component of flagellar type III protein export apparatus**
Takuma Fukumura[1], Fumiaki Makino[1], Takayuki Kato[1], Katsumi Imada[2], Keiichi Namba[1], Tohru Minamino[1] ([1]*Grad. Sch, Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Sci. Osaka Univ*)

2Pos130 **柔軟性に富んだ微小管内 GTPγS-チューブリン分子**
X-ray fiber diffraction analysis revealed a highly flexible state of GTPγS-tubulin in the microtubules
Shinji Kamimura[1], Yosuke Fujita[1], Yuuko Wada[1,2], Tomohiro Shima[3], Yasushi Okada[4], Hiroyuki Iwamoto[5] ([1]*Dept. Biol. Sci., Chuo Univ.*, [2]*Inst. Global Leader.*, [3]*Dept. Biol. Sci., Grad. Sch. Science, Uni. Tokyo*, [4]*QBiC, Riken*, [5]*JASRI, SPring-8*)

2Pos131 **細胞ネットワーク計測のための拡張可能なモジュール型多電極細胞外電位計測システムの開発**
Development of flexible expandable on-chip multi electrode array system for cell-network measurement
Fumimasa Nomura, Akihiro Hattori, Kenji Matsuura, Hiromi Kurotobi, Masao Odaka, Hyonchol Kim, Hideyuki Terazono, Kenji Yasuda (*Institute of Biomaterials and Bioengineering, Tokyo Medical and Dental University*)

2Pos132 **細胞性粘菌の集団運動における細胞極性の動態**
The dynamics of cell polarity during collective migration of *Dictyostelium* cells
Taihei Fujimori[1], Akihiko Nakajima[1,2], Satoshi Sawai[1,2] ([1]*Grad. Sch. Arts & Sci., Univ. Tokyo*, [2]*Res. Ctr. Complex Syst. Biol., Univ. Tokyo*)

2Pos133 **1 細胞分泌実時間イメージングの並列測定プラットフォーム**
Parallel measurement platform for real-time single-cell secretion imaging
Kaede Miyata[1], Yoshitaka Shirasaki[1,2], Nobutake Suzuki[1,2], Mai Yamagishi[1,2], Sotaro Uemura[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*IMS, RIKEN*)

2Pos134 **三次元で見た ATP 依存的な繊毛運動の Ca^{2+} による制御**
Regulation of ATP-dependent ciliary motility by Ca^{2+} observed in three dimensions
Toshihito Iwase[1], Masaaki Suegara[1], Rinako Nakayama[2,5], Takanobu A Katoh[3], Mitsutoshi Setou[4], Takayuki Nishizaka[3], Koji Ikegami[4], Tomoko Masaike[1,2,5] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [2]*Res. Inst. for Sci. and Tech., Tokyo Univ. of Sci.*, [3]*Dept. Phys., Gakushuin Univ.*, [4]*Dept. Cell Biol. and Anat., Hamamatsu Univ. Sch. Med.*, [5]*PRESTO, JST*)

2Pos135 **サルモネラべん毛モーターにおける MotA Met-206 の役割**
Role of MotA Met-206 in *Salmonella* flagellar motor
Kodai Oono[1], Shuichi Nakamura[1], Fumio Hayashi[2], Kenji Oosawa[3], Seishi Kudo[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Instrumental Analysis Center., Gunma Univ.*, [3]*Div. Mol. Sci., Fac. Sci. and Tech, Gunma Univ.*)

2Pos136 **周期的伸展刺激に対する心筋細胞の形態や配向性の変化**
Morphological and orientation change of cardiomyocytes to cyclic stretch stimulation
Chiho Nihei, Tomoyuki Kaneko (*LaRC, Dept. Frontier Biosci., Hosei Univ.*)

2Pos137 **原子間力顕微鏡による肝細胞共培養系の力学特性解析**
Mechanical analysis of hepatocyte coculture system using atomic force microscopy
Ryosuke Tanaka[1], Yoshikatsu Akiyama[2], Jun Kobayashi[2], Masayuki Yamato[2], Takaharu Okajima[1] ([1]*Grad. Sch. Info. Tech., Hokkaido Univ*, [2]*Inst. Adv.BioMed. Eng. Sci., Tokyo Women's Medical Univ*)

2Pos138 **アクチンストレスファイバー内ミオシンのダイナミクス**
Dynamics of nonmuscle myosin molecules in actin stress fibers
Tsubasa S. Matsui, Tomoya Ikeda, Shinji Deguchi (*Dept. Nanoparm. Sci., NITECH*)

2Pos139 **海洋性ビブリオ菌べん毛モーター FliL 蛋白質の生物物理性質と機能解析**
Analysis of function and biophysical properties of marine *Vibrio* FliL in the flagellar motor
Ananthanarayanan Kumar, **Yuuki Nishino**, Shiwei Zhu, Mayuko Sakuma, Seiji Kojima, Michio Homma (*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*)

2Pos140 **ビブリオ菌べん毛モーター MS リング構成因子 FliF の精製タンパク質会合状態**
Oligomeric states of purified FliF, a MS-ring component of Vibrio flagellar motor
Erika Yamaguchi, Seiji Kojima, Michio Homma (*Div. Bio. Sci., Grad. Sch. Sci., Nagoya Univ.*)

2Pos141 **細胞周期での細胞の大きさや形から得られる新たな情報**
New obvious information obtained from the cell size and shape during mitosis cycle
Rina Nagai[1,2], Keisuke Ohta[1,3], Takako Ichinose M.[1,2], Hikari Mori[1,2], **Atsuko Iwane H.**[1,2] ([1]*Cell Field Struc., QBiC, Riken*, [2]*Spec. Res. Promot. Group, Grad. Sch. Fronti., Biosci., Osaka Univ.*, [3]*Anatomy, Med., Kurume Univ.*)

生体膜･人工膜：構造･特性 / Biological & Artificial membrane: Structure & Property

2Pos142 **Bombinin H2 および H4 の細菌模倣膜に対する相互作用の解析**
Analysis of interaction of Bombinin H2 and H4 with the mimetic bacterial membrane
Shiho Kaneda, Yuki Kitahashi, Akira Naito, Izuru Kawamura (*Graduate School of Engineering, Yokohama National University*)

2Pos143 **FTIR-ATR プリズム上に作製したセラミド/ステロール混合膜への重水透過性**
Heavy water permeation into the ceramide/sterol mixed film prepared on an FTIR-ATR prism
Kohei Oka, Satoru Kato (*Grad. Sch. Sci & Tech., Univ. Kwansei Gakuin*)

2Pos144 **セラミドホスホエタノールアミンはマイクロメートルサイズのらせん構造をつくる**
Microstructural Polymorphism of Ceramide Phosphoethanolamine
Takehiko Inaba, Motohide Murate, Yan-Fen Lee, Francoise Hullin-Matsuda, Peter Greimel, Toshihide Kobayashi (*RIKEN Lipid biology laboratory*)

2Pos145 **DNA 構造の自己組織化によるマイクロサイズカプセルの形成**
Microsize capsule formed by self-assembly of DNA structures
Daisuke Ishikawa, Masamune Morita, Masahiro Takinoue (*Interdisciplinary Grad. Sch. Sci. and Eng., Tokyo Tech*)

2Pos146 **細胞膜へのぬれ性がゲル弾性へ及ぼす影響**
How membrane wetting affects elasticity of biopolymer gels in model cells?
Atsushi Sakai, Miho Yanagisawa (*Dept. Appl. Phys.,Tokyo Univ. of Agri. & Tech.*)

2Pos147 **ナノ粒子と質量分析装置を用いて特定タンパク質周辺の脂質を分析する方法の開発について**
Development of a new method to detect lipids surrounding specific proteins using nanoparticles and mass spectrometry
Keiji Seno[1], Yukiyasu Kashiwagi[2], Yumi Yamahama[1] ([1]*Hamamatsu University School of Medicine*, [2]*Osaka Municipal Technical Research Institute (OMTRI)*)

2Pos148 **固体基板が誘起する人工脂質膜の非対称性と異方性**
Substrate-Induced Asymmetry and Anisotropy of Molecular Diffusion in Artificial Lipid Membranes
Toshinori Motegi[1], Kenji Yamazaki[2], Toshio Ogino[3,4], Ryugo Tero[1,4,5] ([1]*EIIRIS, Toyohashi Univ. Tech.*, [2]*Grad. Sch. Eng., Hokkaido Univ.*, [3]*Grad. Sch. Eng., Yokohama Nat. Univ.*, [4]*JST-CREST*, [5]*Dep. Environ. Life Sci., Toyohashi Univ. Tech.*)

2Pos149 **ヌクレオチドに誘起される巨大細胞膜小胞の形態変化**
Nucleotides induce morphological change of giant plasma membrane vesicles
Shun Wakamatsu[1], Takashi Okuno[2] ([1]*Grad. Sch. Sci. Eng., Univ. Yamagata*, [2]*Fac. Sci., Univ. Yamagata*)

生体膜･人工膜：動態 / Biological & Artificial membrane: Dynamics

2Pos150 **単一 GUV 法を用いた細胞透過ペプチド・ポリアルギニンの脂質膜ベシクルへの侵入の研究**
Investigation of the Entry of Cell-Penetrating Peptide, Polyarginine into a Vesicle of Lipid Membrane Using the Single GUV Method
Sabrina Sharmin[1], Md. Zahidul Islam[1], Hideo Dohra[2], Masahito Yamazaki[3,4] ([1]*Int. Biosci., Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Green Sci. Tech., Shizuoka University*, [3]*Res. Inst. Electronics, Shizuoka Univ.*, [4]*Dept. Phys., Grad. Sch. Sci., Shizuoka Univ.*)

2Pos151 **低い pH が誘起する DOPS/MO 膜の液晶相からキュービック相への相転移に対する温度効果**
The Effect of Temperature on the Low pH-Induced Lamellar to Bicontinuous Cubic Phase Transition in DOPS/MO
Toshihiko Oka[1,2], Takahiro Saiki[1], Jahangir Md. Alam[2], **Masahito Yamazaki**[1,2] ([1]*Dept. Phys., Grad. Sch. Sci., Shizuoka Univ.*, [2]*Res. Inst. Electronics, Shizuoka Univ.*)

2Pos152 **Effects of Cholesterol on the Entry of Cell-Penetrating Peptide Transportan 10 (TP10) into a Single Vesicle of Lipid Membranes**
Md. Zahidul Islam[1], Sabrina Sharmin[1], Masahito Yamazaki[1,2] ([1]*Int. Biosci., Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Electronics, Shizuoka Univ.*)

2Pos153 **アクチン骨格様の DNA ゲル薄膜で支持されたリポソームの構築**
Liposomes with skeleton network of self-assembled DNA gel mimicking actin cortex
Chikako Kurokawa[1], Kei Fujiwara[2], Masamune Morita[3], Ibuki Kawamata[4], Satoshi Murata[4], Masahiro Takinoue[3], Miho Yanagisawa[1] ([1]*Dept. Appl. Phys., Tokyo Univ. of Agri. & Tech.*, [2]*Keio Univ.*, [3]*Tokyo Inst. Technol.*, [4]*Tohoku Univ.*)

2Pos154 **支持脂質二重膜へのプロテオリポソーム再構成過程の観察**
Observation of reconstitution process of proteoliposome into supported lipid bilayer
Kohei Fukumoto[1], Yutaka Ishinari[2,3], Ayumi Hirano-Iwata[2,3], Michio Niwano[3,4], Ryugo Tero[1,3,5] ([1]*Dept. Environment. Life Sci., Toyohashi Univ. Tech.*, [2]*Grad. Sch. Biomed. Eng., Tohoku Univ.*, [3]*CREST, JST*, [4]*RIEC, Tohoku Univ.*, [5]*EIIRIS, Toyohashi Univ. Tech.*)

2Pos155 **グラフェン酸化物が誘起する中性リン脂質膜からなる巨大単一膜ベシクルの形状変化**
Graphene oxide induced structural transformation of single giant unilamellar vesicles of phosphatidylcholine membranes
Gento Nakagawa[1], Yoshiaki Okamoto[2], Ryugo Tero[2], Yukihiro Tamba[1] ([1]*Suzuka Natl. Coll. Tech.*, [2]*Toyohashi Univ. Tech.*)

生体膜･人工膜：興奮･チャネル / Biological & Artificial membrane: Excitation & Channels

2Pos156 **KcsA チャネルの細胞内ドメインによる不活性化への影響**
The cytoplasmic domain regulates inactivation in the KcsA channel
Minako Hirano[1], Yukiko Onishi[2], Toru Ide[3] ([1]*GPI*, [2]*RIKEN, Qbic*, [3]*Okayama Univ.*)

2Pos157 **微小な液滴接触膜の形成とイオンチャネル機能解析への応用**
The formation of a contact bubble bilayer and its application to the functional analysis of ion channels
Masayuki Iwamoto, Shigetoshi Oiki (*Dept. Mol. Physiol. Biophys., Univ. Fukui Facult. Med. Sci.*)

2Pos158 **分子動力学で明らかにされる細菌機械受容チャネル MscL の N 末端領域の張力センサーとしての重要性**
How Important is the N-Terminal Domain of Bacterial Mechanosensitive Channel MscL for Sensing Membrane Tension: Molecular Dynamics Study
Yasuyuki Sawada[1], Masahiro Sokabe[2] ([1]*Dept. Physiol. Nagoya Univ. Grad. Sch. Med.*, [2]*Mechanobiology Lab. Nagoya Univ. Grad. Sch. Med.*)

生体膜･人工膜：輸送 / Biological & Artificial membrane: Transport

2Pos159 **心筋 Na/Ca 交換の PMCA とカルシウムホールによる調節**
Regulation of cardiac Na/Ca exchanger by PMCA via "calcium holes"
Takao Shioya (*Dept. Physiol. Fac. Med. Saga Univ.*)

2Pos160 **細胞-GUV 電気融合による μm スケールの人工物導入について**
Electrofusion of Cell-GUV enables um-sized artificial objects transfer into live cells
Akira C. Saito[1], Toshihiko Ogura[2], Satoshi Murata[1], Shin-ichiro Nomura[1] ([1]*Department of Bioengi. and Robo. Tohoku Univ.*, [2]*Depart. of Develo. of Neurobiolo. (IDAC). Tohoku. Univ.*)

化学受容 / Chemoreception

2Pos161 **コレラ菌全走化性受容体ホモログの Che システム帰属**
Che system assignment of all chemoreceptor homologs in *Vibrio cholerae*
So-ichiro Nishiyama[1,2], Akihiro Hyakutake[3], Noriko Nishioka[3], Michio Homma[3], Ikuro Kawagishi[1] ([1]*Dept. Frontier Biosci., Hosei Univ.*, [2]*Res. Cen. Micro-Nano Tech., Hosei Univ.*, [3]*Div. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

神経科学･感覚 / Neuroscience & Sensory systems

2Pos162 **新規光応答性 CaMKII の単一スパイン内光操作**
Optogenetic manipulation of photo-activatable CaMKIIα in individual dendritic spines of neuron
Akihiro Shibata[1,2], Hideji Murakoshi[1] ([1]*National Institute for Physiological Science*, [2]*JSPS Research Fellow*)

2Pos163 **ナメクジ嗅覚中枢の培養ニューロンにおける神経振動ネットワークの再形成とアセチルコリン／ヒスタミンによる自発的神経活動の調節**

Oscillatory network formation and cholinergic/histaminergic activity in the cultured olfactory neurons in the slug

Suguru Kobayashi, Asuka Kobayashi (*Kagawa Schl Pharmaceut Sci, Tokushima Bunri Univ*)

2Pos164 **海馬で合成される男性・女性ホルモンによる記憶シナプスの急性制御の解析**

Rapid modulation of memory-related synapses by locally synthesized sex-hormones in the hippocampus

Suguru Kawato[1,2], Asami Kato[2], Yasushi Hojo[1] ([1]*Juntendo Univ, Med*, [2]*Univ. Tokyo*)

神経回路･情報処理 / Neuronal circuit & Information processing

2Pos165 **ミミズ非連合学習における 5-HT/NO/cGMP シグナルの役割**

Role of 5-HT/NO/cGMP signaling cascade in non-associative learning of earthworm

Yoshiichiro Kitamura[1], Hitoshi Aonuma[2], Hiroto Ogawa[3], Kotaro Oka[4] ([1]*Dept Math Sci Phys, Kanto Gakuin Univ*, [2]*Res Inst Elect Sci, Hokkaido Univ*, [3]*Dept Biol Sci, Hokkaido Univ*, [4]*Dept Biosci Info, Keio Univ*)

2Pos166 **柔らかい有機電極を用いる脳活動の同時多点測定**

Multi-site recording of brain activity using flexible organic electrodes

Satoshi Watanabe, Hideyuki Takahashi, Keiichi Torimitsu (*Grad. Sch. Eng., Tohoku Univ.*)

行動 / Behavior

2Pos167 **2-AG 分解酵素モノアシルグリセロールリパーゼ欠損マウスにおける課題依存性学習障害**

Task-specific impairment of hippocampus-dependent learning in mice deficient in monoacylglycerol lipase

Yasushi Kishimoto[1], Barbara Cagniard[2], Maya Yamazaki[3], Junko Nakayama[1], Kenji Sakimura[3], Yutaka Kirino[1], Masanobu Kano[2] ([1]*Department of Neurobiophysics, Kagawa School of Pharmaceutical Sciences, Tokushima Bunri University*, [2]*Department of Neurophysiology, Graduate School of Medicine, University of Tokyo*, [3]*Department of Cellular Neurobiology, Brain Research Institute, Niigata University*)

光生物：視覚･光受容 / Photobiology: Vision & Photoreception

2Pos168 **X-ray Crystal Structure of TR: Implications for High Thermal Stability and High-Performance Optogenetic Availability**

Takashi Tsukamoto[1], Kenji Mizutani[2], Megumi Takahashi[3], Taisuke Hasegawa[4], Naoki Hashimoto[2], Shigehiko Hayashi[4], Shin Takagi[3], Takeshi Murata[2], Yuki Sudo[1] ([1]*Okayama University*, [2]*Chiba University*, [3]*Nagoya University*, [4]*Kyoto University*)

2Pos169 **Synechocystis sp. PCC 7509 由来の新規光駆動アニオンポンプの機能解析**

Functional studies on a light-driven anion pump from Synechocystis sp. PCC 7509

Akiko Niho[1], Susumu Yoshizawa[2], Yu Nakajima[2], Takashi Tsukamoto[1,3], Yuki Sudo[1,3] ([1]*Dept. Pharm. Sci., Okayama Univ.*, [2]*Atm. Ocean Res. Inst., Univ. Tokyo*, [3]*Grad. Sch. of Med. Dent. Pharm. Sci, Okayama Univ.*)

2Pos170 **プロテオロドプシンのアルカリ性条件下における光誘起プロトン移動の pH 依存性**

pH dependence of the photoinduced proton transfer in proteorhodopsin under alkaline conditions

Jun Tamogami[1], Keitaro Sato[2], Sukuna Kurokawa[2], Takumi Yamada[2], Toshifumi Nara[1], Makoto Demura[3], Takashi Kikukawa[3], Eiro Muneyuki[2], Naoki Kamo[3] ([1]*College Pharm. Sci., Matsuyama Univ.*, [2]*Grad. Sci and Eng., Chuo Univ.*, [3]*Fac. Adv. Life. Sci., Hokkaido Univ.*)

2Pos171 **アフリカツメガエル由来(6-4)光回復酵素の変異解析**

Mutational analyses of Xenopus laevis (6-4) photolyase

Kohei Shimizu[1], Takahiro Yumiba[1], Tomoko Ishikawa[2], Takeshi Todo[2], Junpei Yamamoto[1], Shigenori Iwai[1] ([1]*Grad. Sch. Eng. Sci., Univ. Osaka*, [2]*Grad. Sch. Med., Univ. Osaka*)

2Pos172 **量子化学計算を用いたプロテオロドプシン L105 変異体の波長シフトの解析**

Theoretical analysis of the color-tuning mechanism of mutations at Leu105 in Green-Light Absorbing Proteorhodopsin

Kaichi Yanagi, Hiroshi C. Watanabe, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech*)

2Pos173 **Photoactive Yellow Protein におけるアルギニン 52 のプロトン化状態**

Protonation State of Arginine 52 in Photoactive Yellow Protein

Kento Yonezawa[1], Hironari Kamikubo[1], Yusuke Kanematsu[2], Yoichi Yamazaki[1], Masanori Tachikawa[2], Mikio Kataoka[1] ([1]*Grad. Sch. Mat. Sci NAIST*, [2]*Graduate School of Nanobioscience, Yokohama City Univ*)

2Pos174 ***Rhodobacter capsulatus* 由来 Photoactive Yellow Protein の相互作用における β-scaffold 部位の役割**

Analysis of interaction sites on β-scaffold region of *Rhodobacter capsulatus* Photoactive Yellow Protein

Yoichi Yamazaki, Atsuhiro Kawamura, Mikio Kataoka, Hironari Kamikubo (*Materials Science NAIST*)

2Pos175 **ナトリウムポンプロドプシンの低温赤外分光研究**

Low-temperature FTIR Study of Sodium Pumping Rhodopsin

Shota Ito[1], Shinya Sugita[1], Rei Yoshizumi-Abe[1], Keiichi Inoue[1,2], Tatsuya Iwata[1], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Prest, JST*)

2Pos176 **光駆動ナトリウムポンプ KR2 のポンプスイッチにおける Asn112 の役割**

Role of Asn112 for transport activity by a light-driven sodium ion pump

Rei Abe-Yoshziumi[1], Keiichi Inoue[1,2], Hideaki Kato[3], Osamu Nureki[4], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*, [3]*Stanford University Medical School*, [4]*Grad. Sch. of Sci., Univ. of Tokyo*)

2Pos177 **部位特異的変異体を用いた bZIP モジュールである Photozipper の戻り反応の評価**
Dark regeneration kinetics of site-directed mutants of bZIP module, Photozipper
Yuki Yabe, osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

光生物：光合成 / Photobiology: Photosynthesis

2Pos178 **酸素発生複合体におけるＳ４状態での３重項酸素発生に関する理論的研究**
Theoretical study on evolution of triplet oxygen molecule at the S4 state of oxygen evolution complex
Yasunori Yoshioka[1], Tomoya Ichino[2] ([1]*Mie University*, [2]*Hokkaido University*)

2Pos179 **好熱性紅色光合成細菌 Thermochromatium tepidum 由来 LH1-RC 複合体におけるキノン分子の検出**
Detection of quinone molecules in the LH1-RC complex from the thermophilic purple photosynthetic bacterium Thermochromatium tepidum
Mari Matsuzaki[1], Yuki Yura[1], Takashi Ohno[1], Seiu Otomo[2], Yukihiro Kimura[1] ([1]*Grad. Sch. Agri. Sci., Kobe Univ.*, [2]*Fac. Sci., Ibaraki Univ.*)

2Pos180 **NMR study of the interaction sites on the two Fd isoforms for photosynthetic protein complexes**
Risa Mutoh[1], Yuko Misumi[1], Hisako Kubota-Kawai[2], Ryutaro Tokutsu[2], Takahisa Ikegami[3], Hippler Michael[4], Jun Minagawa[2], Genji Kurisu[1] ([1]*Inst. Prot. Res., Osaka Univ.*, [2]*National Inst. Basic Biol.*, [3]*Dep. Med. Life Sci., Yokohama City Univ.*, [4]*Inst. Plant Biol. and Biotech., Univ. of Munster*)

2Pos181 **Fluorescence spectroscopy of single Photosystem I at liquid nitrogen temperatures**
Ting Du (*Tohoku University*)

2Pos182 **励起子緩和過程の摂動論に対する新たな試み**
New approach of perturbative study in exciton relaxation process
Akihiro Kimura (*Department of Physics, Graduate School of Science, Nagoya University*)

2Pos183 **ENDOR studies on biochemical modification on calcium site of the Mn cluster in photosystem II**
Hiroki Nagashima[1], Yoshiki Nakajima[2], Jian-Ren Shen[2], Hiroyuki Mino[1] ([1]*Grad. Sch, Sci., Nagoya Univ.*, [2]*Grad. Sch. Nat. Sci. and Tech./Fac. Sci., Okayama Univ.,*)

放射線生物学･活性酸素 / Radiobiology & Active oxygen

2Pos184 **プリオンペプチドと二価金属錯体結合によってひきおこされるレドックス不均衡**
Redox imbalance induced by coordination of metals in prion peptide
Shinnosuke Kondo, Masahiro Yagi, Wakako Hiraoka (*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*)

2Pos185 **超音波による CMNB ケージ基解離と脂肪酸分解の比較**
Comparison between CMNB-caged moiety and fatty acid on molecular scission induced by ultrasound
Kengo Takei[1], Haruko Koura[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Master. & Human Env. Sci., Shonan Inst. of Tech.*)

生命の起源･進化 / Origin of life & Evolution

2Pos186 **Self-Emergent Cell-Sized Microsphere Entrapping DNA in a Crowding Binary Polymer Solution**
Naoki Nakatani[1], Kanta Tsumoto[2], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life Medical Sci., Doshisha Univ.*, [2]*Grad. Sch. Engineering, Mie Univ.*)

ゲノム生物：ゲノム解析 / Genome biology: Genome analysis

2Pos187 **Culture-independent method for identifying microbial enzyme-encoding genes based on activity-driven single cell genomics**
Kazuki Nakamura[1], Ryo Iizuka[1], Takao Yoshida[2], Yuji Hatada[2], Yoshihiro Takaki[2], Shinro Nishi[2], Ayaka Iguchi[3], Dong Hyun Yoon[3], Tetsushi Sekiguchi[3], Shuichi Shoji[3], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*JAMSTEC*, [3]*Major in Nanosci. and Nanoeng., Waseda Univ.*)

ゲノム生物：ゲノム構造 / Genome biology: Genome structure

2Pos188 **ヒト間期核の全ゲノム動力学シミュレーション**
Genome-wide chromatin dynamics simulation of human interphase nucleus
Shin Fujishiro, Naoko Tokuda, Masaki Sasai (*Grad. Sch. Eng., Nagoya Univ.*)

バイオインフォマティクス：ゲノム構造 / Bioinformatics: Structural genomics

2Pos189 **リガンド結合による構造変化がもたらす STING シグナル伝達系への影響**
Ligand-induced conformational changes in STING are essential for its signal transduction
Yuko Tsuchiya[1], Kenji Mizuguchi[2] ([1]*Institute for Protein Research*, [2]*National institutes of Biomedical Innovation, Health and Nutrition*)

2Pos190 **ベクトルマッチアルゴリズムによるタンパク質―タンパク質ドッキングポーズを評価するための統計アミノ酸ペアポテンシャルの開発**
A statistical amino acid pair potential to re-rank protein-protein docking poses predicted by a vector match algorithm
Atsushi Hijikata, Masafumi Shionyu, Tsuyoshi Shirai (*Facl. Biosci., Nagahama Inst. Bio-Sci. Tech.*)

2Pos191 **埋もれた極性残基の進化的保存**
Evolutionary conservation of buried polar residues
Matsuyuki Shirota[1,2,3] ([1]*Grad Sch Med, Tohoku Univ*, [2]*ToMMo, Tohoku Univ*, [3]*GSIS, Tohoku Univ*)

2Pos192 **高い配列相同性を持ちながら異なる立体構造を持つタンパク質のアミノ酸配列と立体構造に基づく解析**
The differences in 3D structures of proteins with high sequence identity. Analyses of amino acid sequences and 3D structures
Kohei Ohnishi, Masanari Matsuoka, Masatake Sugita, Takeshi Kikuchi (*Ritsumeikan Univ.*)

バイオインフォマティクス：ゲノム機能 / Bioinformatics: Functional genomics

2Pos193 **機能未知スプライシングアイソフォームの機能予測**
Function prediction of uncharacterized splicing isoforms
Masafumi Shionyu (*Fac. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*)

バイオインフォマティクス：分子進化 / Bioinformatics: Molecular evolution

2Pos194 **リゾチームスーパーファミリータンパク質のフォールディングユニットの頑健性.タンパク質機能とフォールディング機構との関係**
Robustness of folding units in lysozyme superfamily proteins during evolution, relationship between functions and folding mechanisms
Takuto Nakashima, Michirou Kabata (*Ritsumeikan Univ.*)

数理生物 / Mathematical biology

2Pos195 **構造変化を伴う高分子の反応拡散系―構造と機能のクロストーク**
Reaction-Diffusion Systems with Polymers Consisting of Mechanical Units: Crosstalk between the Structure and Function
Yuichi Togashi (*RcMcD, Hiroshima Univ.*)

2Pos196 **ミドリゾウリムシの細胞内共生における共生藻の単純な維持機構**
Maintenance of algal endosymbionts in *Paramecium bursaria*: a simple model based on population dynamics
Sosuke Iwai, Kenji Fujiwara, Takuro Tamura (*Faculty of Education, Hirosaki Univ.*)

2Pos197 **遺伝子発現振動系のダイナミクス低減**
Dynamical Reduction Approach reveals Oscillating Gene Expressions
Ikuhiro Yamaguchi[1], Yutaro Ogawa[2], Akihiko Akao[2], Yuki Shimono[2], Yasuhiko Jimbo[3], Kiyoshi Kotani[4] ([1]*Grad. Edu. Univ. Tokyo*, [2]*Front. Sci. Univ. Tokyo*, [3]*Eng. Univ. Tokyo*, [4]*RCAST Univ. Tokyo*)

2Pos198 **複雑反応ネットワークに埋め込まれた時間階層構造の解読**
Deciphering timescale hierarchy encoded in complex reaction networks
Yutaka Nagahata[1], Satoshi Maeda[2], Hiroshi Teramoto[1,3], Chun-Biu Li[2,3], Takashi Horiyama[4], Tetsuya Taketsugu[2], Tamiki Komatsuzaki[1,3] ([1]*Graducate School of Life Science, Hokkaido Univ.*, [2]*Faculty of Science, Hokkaido Univ.*, [3]*Research Institute for Electronic Science, Hokkaido Univ.*, [4]*Graduate School of Science and Engineering, Saitama Univ.*)

2Pos199 **Interdomain communication as the mechanism of correlation between circadian oscillation of KaiC phosphorylation and ATPase activity**
Shota Hashimoto, Das Sumita, Masaki Sasai, Tomoki P. Terada (*Grad. Sch. Eng., Nagoya Univ.*)

非平衡･生体リズム / Nonequilibrium state & Biological rhythm

2Pos200 **人工 RNA 自己複製システムにおける宿主 RNA と寄生体 RNA の振動ダイナミクスと進化**
Oscillating population dynamics and evolution of artificial Host-Parasite replication system in micro compartment
Norikazu Ichihashi[1], Yohsuke Bansho[2], Tetsuya Yomo[1,2] ([1]*Osaka Univ, IST*, [2]*Osaka Univ, FBS*)

2Pos201 **微小液滴を用いた非線形化学反応間の相互作用の制御**
Control of the interaction among nonlinear chemical reactions based on microdroplets
Tomoya Okuaki[1], Haruka Sugiura[1], Ryuji Kawano[2], Masahiro Takinoue[1] ([1]*Dept. Comput. Intell. Syst. Sci., Tokyo Tech*, [2]*Div. Biotech. & life Sci., TUAT*)

計測 / Measurements

2Pos202 **細胞チップの電気生理学的特性評価のためのインピーダンス／細胞外電位計測システムの開発**
Development of impedance measurement system for identification of cells
Kenji Matsuura[1], Fumimasa Nomura[2], Akihiro Hattori[1], Hiromi Kurotobi[2], Masao Odaka[1], Hyonchol Kim[1], Hideyuki Terazono[2], Kenji Yasuda[1,2] ([1]*Kanagawa Academy of Science and Technology*, [2]*Tokyo Medical and Dental Univ.*)

2Pos203 **オンチップマルチイメージングセルシステムを用いた血液中の単一がん細胞を認識するためのイメージングバイオマーカー認識方法の評価**
Evaluation of imaging biomarkers for identification of single cancer cells in blood by on-chip multi imaging cell system
Masao Odaka[1,2], Hyonchol Kim[1,2], Mathias Girault[1], Akihiro Hattori[1], Hideyuki Terazono[1,2], Kenji Matsuura[1], Fumimasa Nomura[2], Kenji Yasuda[1,2] ([1]*KAST*, [2]*IBB, TMDU*)

2Pos204 **FCS による HbA1c の計測**

HbA1c measurement using FCS

Atsushi Matsuo, Yasutomo Nomura, Kyohei Maruyama, Kyohei Nakayama, Mayuka Chiba (*Maebashi Institute of Technology*)

2Pos205 **生細胞における 1 分子内在性 mRNA イメージングのためのアンチセンスプローブの設計と評価**

Design and evaluation of potent antisense probes for imaging individual endogenous mRNA in live cells

Shunsuke Takeda[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

2Pos206 **蛍光偏光相関分光法による蛍光タンパク質の回転拡散の研究**

Study of rotational diffusion of fluorescent proteins using polarization dependent fluorescence correlation spectroscopy (pol-FCS)

Makoto Oura[1], Johtaro Yamamoto[2], Masataka Kinjo[2] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Advanced Life Sci., Hokkaido Univ.*)

2Pos207 **電子線の動電現象による単一接着性細胞への局所的な染色液導入の観察**

Observation of Local Dye Inflow into Single Adherent Cells induced by Electrokinetic Phenomena of Electron Beam

Moto Yoshioka, Hiroki Miyazako, Akira Wagatsuma, Kunihiko Mabuchi, Takayuki Hoshino (*Grad. Sch. IST., Univ. Tokyo*)

バイオイメージング / Bioimaging

2Pos208 **結合速度と光感受性を改良した蛍光 ATP バイオセンサー**

Improvement of binding speed and photostability of fluorescent ATP biosensor for extracellular ATP imaging

Hiromi Imamura, Sui Nishiyama, Ryuta Iwakiri, Akira Kakizuka (*Grad. Biostudies, Kyoto Univ.*)

2Pos209 **高速 AFM による DNA ジャイレースのダイナミクスの直接観察**

Direct observation of dynamic action in DNA gyrase by high-speed AFM

Daisuke Noshiro[1], Noriyuki Kodera[2,3], Toshio Ando[1,2,4] ([1]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [2]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [3]*PRESTO, JST*, [4]*CREST, JST*)

2Pos210 **電子顕微鏡画像処理及び画像解析のための Eos/PIONE の開発**

Development of Eos and PIONE for Image Processing and Analysis of Electron Micrographs

Takuo Yasunaga[1], Keita Yamaguchi[2], Takafumi Tsukamoto[1] ([1]*Dept. of Biosci. and Bioinfo., Sch. of Comp. Sci. and Sys. Eng., Kyushu Inst. Tech.*, [2]*Nau Data Inc.*)

2Pos211 **ラスター画像相互相関分光法による生細胞内外来 DNA 分解活性の時空間的可視化**

Raster image cross-correlation method for spatiotemporal visualization of intracellular degradation activities against exogenous DNAs

Akira Sasaki[1,2], Johtaro Yamamoto[3], Takashi Jin[2], Masataka Kinjo[3] ([1]*BMRI, AIST*, [2]*QBiC, Riken*, [3]*Faculty of Adv. Life Sci., Hokkaido Univ.*)

2Pos212 **Novel green fluorescent protein from Olindias formosa with exceptional pH stability**

Hajime Shinoda[1], Yuanqing Ma[2], Tomoki Matsuda[1,3], Takeharu Nagai[1,3] ([1]*Grad. Sch. Eng., Univ. Osaka*, [2]*Univ. Western Sydney*, [3]*ISIR, Univ. Osaka*)

2Pos213 **赤外超解像顕微鏡法による毛髪 α-ケラチンの分子配向イメージング -振動モード毎の偏光依存性測定-**

Orientation-sensitive IR super-resolution imaging of human hair α-keratins -Polarization dependency measurements-

Kohei Ushio, Yukihisa Watase, Masaaki Fujii, Makoto Sakai (*Tokyo Institute of Technology*)

2Pos214 **１分子イメージングによる転写伸長因子 NELF と DSIF のダイナミクス定量解析**

Quantitative analysis of dynamics of negative elongation factor NELF and DSIF by single molecule imaging

Daichi Ikeda, Yuma Ito, Makio Tokunaga, Kumiko Sakata-Sogawa (*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*)

2Pos215 **ストレス顆粒内一分子 mRNA の超解像イメージング**

Super-resolution imaging of single mRNA in stress granules

Yuki Suzuki[1], Ko Sugawara[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharma., Univ. Tokyo*, [2]*JST, PRESTO*)

2Pos216 **ディフュージョンマップ法を用いたタンパク質位相回復像の分類**

Classification of phase-retrieved projection electron density maps of a protein using the diffusion-map method

Takashi Yoshidome[1], Oroguchi Tomotaka[2,3], Masayoshi Nakasako[2,3], Mitsunori Ikeguchi[4] ([1]*Department of Applied Physics, School of Engineering, Tohoku University, Japan*, [2]*Department of Physics, Faculty of Science and Technology, Keio University*, [3]*Research Infrastructure Group, Advanced Photon Technology Division, RIKEN Harima Institute, Japan*, [4]*Graduate School of Medical Life Science, Yokohama City University, Japan*)

2Pos217 **ラスター画像相関分光法(RICS)による生細胞内の DNA 分解過程の時空間解析**

Spatiotemporal analysis of exogenous DNA degradation in living cells by raster image correlation spectroscopy (RICS)

Takashi Horio[1], Johtaro Yamamoto[2], Akira Sasaki[3], Masataka Kinjo[2] ([1]*Lab. Mol. Cell Dynamics, Grad. Life Sci., Hokkaido Univ.*, [2]*Lab. Mol. Cell Dynamics, Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*Biomedical Research Inst., AIST.*)

2Pos218 **コヒーレント X 線回折イメージング法に向けたフーリエ変換ホログラフィー法による初期位相決定法の開発**

Application of Fourier transform holography to initial phasing in coherent X-ray diffraction imaging

Yuki Takayama[1], Yayoi Inui[2], Yuki Sekiguchi[1,3], Sachihiro Matsunaga[2], Masayoshi Nakasako[1,3], Koji Yonekura[1] ([1]*RIKEN SPring-8 Center*, [2]*Sci. Tech., Tokyo Univ. Sci.*, [3]*Sci. Tech., Keio Univ.*)

バイオエンジニアリング / Bioengineering

2Pos219 **血管内皮細胞の自発的管形成能力を利用した in vitro 組織培養システムの構築**
In vitro perfused tissue culture system using spontaneous vascular formation of endothelial cell
Yuji Nashimoto[1,4], Akiko Nakamasu[2,4], Hisako Imamura[2,4], Hidetoshi Kotera[1], Koichi Nishiyama[3,4], Takashi Miura[2,4], Ryuji Yokokawa[1,4] ([1]*Kyoto University, Graduate School of Engineering*, [2]*Kyusyu university, Graduate School of Medical Sciences*, [3]*Kumamoto University, Graduate School of Medical Sciences*, [4]*JST, CREST*)

2Pos220 **マイクロ波照射下での酵素反応の促進効果**
Acceleration of Enzymatic Reaction under Microwave Irradiation
Arata Shiraishi[1], Takeo Yoshimura[2], Seiji Higa[1], Hiroya Osoegawa[1], Shokichi Ohuchi[1,3] ([1]*Dept. Lifesci. & Syst. Eng., Kyushu Inst. Tech*, [2]*Dept. Appl. Chem., Tokyo Inst. Tech.*, [3]*Dept. Biosci. & Bioinform., Kyushu Inst. Tech.*)

2Pos221 **マイクロ凹形状構造によるサイズ選択的細胞回収への排除体積効果の寄与**
Contribution of depletion effect to size-specific target cell purification using mirometer-sized concave structures
Hyonchol Kim[1,2], Hideyuki Terazono[2], Hiroyuki Takei[3], Akihiro Hattori[1], Kenji Matsuura[1], Fumimasa Nomura[2], Kenji Yasuda[2] ([1]*KAST*, [2]*Inst. Biomat. Bioeng., Tokyo Med. Dent. Univ.*, [3]*Facul. Life Sci., Toyo Univ.*)

2Pos222 **微生物（大腸菌）を使った化学物質センサーの開発**
Development of chemical substance sensor by using micro organism (*E. coli*)
Hiroto Tanaka[1], Tadashi Matsukawa[1], Yasushi Naruse[2], Yukihiro Tominari[3], Masato Okada[4], Yoshiyuki Sowa[5], Ikuro Kawagishi[5], Hiroaki Kojima[1] ([1]*Bio ICT Lab, NICT*, [2]*CiNet, NICT*, [3]*Nano ICT, NICT*, [4]*Tokyo Univ.*, [5]*Hosei Univ.*)

2Pos223 **リアルタイム局所化学刺激システムの開発と応用**
Development and application of the real-time local chemical stimulation system
Masaru Kojima, Takahiro Motoyoshi, Mitsuhiro Horade, Kazuto Kamiyama, Yasushi Mae, Tatsuo Arai (*Grad. Sch. Eng. Sci., Osaka Univ.*)

2Pos224 **DNA origami を用いた直交性のある転写ナノデバイスの構築**
Rational design of orthogonal gene transcription nano device on DNA origami
Takeya Masubuchi[1], Hisashi Tadakuma[2], Ryo Iizuka[3], Masayuki Endo[2], Takashi Funatsu[3], Hiroshi Sugiyama[2], Yoshie Harada[2], Takuya Ueda[1] ([1]*Grad. Sch. of Frontier Sci., The Univ. of Tokyo*, [2]*iCeMS, Univ. Kyoto*, [3]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

2Pos225 **生体ナノポアと 3-way junction DNA を用いた 1 分子ロジックゲートの構築**
Single molecule logic operations using 3-way junction DNA and biological nanopores
Masayuki Ohara, Ryuji Kawano (*Grad. Sch. Eng., Tokyo Univ. of Agr. and Tech.*)

2Pos226 **近赤外レーザーによる安定な細胞集合体の構築**
Construction of Stable Cellular Assembly with Optical Manipulation
Aoi Yoshida, Shu Hashimoto, Taeko Ohta, Kenichi Yoshikawa (*Doshisha University*)

2Pos227 **マイクロピペットによるリポソーム内への物質移入法の開発**
Development of novel methods for introducing materials into liposomes with micropipettes
Shin Yoshida[1], Fumika Asari[2], Tomoyuki Kaneko[1,2] ([1]*LaRC, Grad. Sci. Eng., Hosei Univ.*, [2]*LaRC, Dept. Frontier Biosci., Hosei Univ.*)

2Pos228 **生化学分析のための電子線による有機カチオン輸送制御**
Transportation Control of Organic Cations Using an Electron-beam for Biochemical Analysis
Hiroki Miyazako[1,2], Kyoko Fujita[3,4], Nobuhumi Nakamura[3,4], Hiroyuki Ohno[3,4], Kunihiko Mabuchi[1], Takayuki Hoshino[1] ([1]*IPC, UTokyo*, [2]*JSPS Research Fellow*, [3]*Dept. Biotech., TUAT*, [4]*Grad. Sch. Eng., TUAT*)

その他 / Miscellaneous topics

2Pos229 **ビリルビンを発色団とする蛍光タンパク質 UnaG のキロオプティカル特性**
Chiroptical Properties of Bilirubin-based Fluorescent Protein UnaG
Togo Shimozawa, Yoh Shitashima, Miyabi Ishida, Toru Asahi (*Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*)

2Pos230 **Measuring *Escherichia coli* flagellar filaments growth in real time using fluorescent microscopy**
Ziyi Zhao[1], Chien-Jung Lo[2], Fan Bai[1] ([1]*Biodynamic Optical Imaging Center, Peking University, Beijing, China*, [2]*Department of Physics and Graduate Institute of Biophysics, National Central University, Jhongli, Taiwan*)

第 3 日目（9 月 15 日（火））／Day 3（Sep. 15 Tue.）プロムナード / Promenade

蛋白質：構造 / Protein: Structure

3Pos001 **Rap1B の構造とコンフォメーションスイッチ機構**
The structure and conformational switching of Rap1B
Hiroki Noguchi, Sam-Yong Park, Jeremy Tame, **Satoru Unzai** (*Yokohama City University, Graduate School of Medical Life Science*)

3Pos002 **完全重水素化 HiPIP の構造と性質**
Structure and characterization of perdeuterated HiPIP
Yuya Hanazono, Kazuki Takeda, Kunio Miki (*Grad. Sch. Sci., Kyoto Univ.*)

3Pos003 **酸化ヌクレオチド分解酵素の高分解能 X 線回折**
High resolution X-ray diffraction study of an enzyme for oxidative nucleotide processing
Keisuke Hirata, Teruya Nakamura, Mami Chirifu, Shinji Ikemizu, **Yuriko Yamagata** (*Grad. Sch. Pharm., Kumamoto Univ*)

3Pos004 **Structural and functional studies on two kinds of perireceptor proteins (PRPs) working in chemoreception**
Xing Li[1], Durige Wen[2], Masaru Hojo[3], Mamiko Ozaki[3], Tatsuo Iwasa[1,4] ([1]*Div. Eng., Muroran Ins. of Tech.*, [2]*Div of Prod Sys Eng., Muroran Ins. of Tech.*, [3]*Dept. Biol., Grad. School Sci., Kobe Univ*, [4]*Cen. Env. Sci. Dis. Mit. Adv. Res., Muroran Ins. of Tech.*)

3Pos005 **ストマチンパートナータンパク質の結晶構造と多量体形成**
Crystal structure of stomatin operon partner protein and formation of multimeric assembly
Hideshi Yokoyama[1], Ikuo Matsui[2] ([1]*Sch. of Pharm. Sci., Univ. of Shizuoka*, [2]*Biomedical Res. Inst., AIST*)

3Pos006 **分子動力学シミュレーションを用いたリガンド-レセプター間の最適解離経路を決定するための新規方法**
A novel method to determine the optimal unbinding path between receptor and ligand using advanced molecular dynamics simulations
Gert-Jan Bekker[1,2], Narutoshi Kamiya[1], Haruki Nakamura[1] ([1]*IPR, Osaka Univ*, [2]*FBS, Osaka Univ*)

3Pos007 **赤痢菌ニードル複合体の極低温電子顕微鏡による構造解析**
CryoEM structural analysis of the needle complex from *Shigella flexneri*
Naoko Kajimiura[1], Fumiaki Makino[1], Takayuki Kato[1], Ariel Blocker[2], Keiichi Namba[1,3] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*Sch. of Cell. & Mol. Med., Univ. of Bristol*, [3]*RIKEN, QBiC*)

3Pos008 **低解像度密度マップへの複数のサブユニットのあてはめ計算 - 実験情報による拘束の利用 -**
Multiple subunit fitting into a low resolution density map using experimental additional restraints
Takeshi Kawabata, Hirofumi Suzuki, Haruki Nakamura (*Inst. Prot.Res., Osaka Univ.*)

3Pos009 **SAAP 力場を用いたシニョリンのレプリカ交換モンテカルロシミュレーション**
Replica-exchange Monte Carlo simulation of chignolin using SAAP force field
Toshiki Suzuki, Taku Shimosato, Natsuki Babe, Toshiya Minezaki, Michio Iwaoka (*School Sci., Tokai Univ.*)

3Pos010 **全原子 MD シミュレーションによるアラニンスキャニング変異を導入したアミロイド形成ペプチドの凝集解析**
All atom molecular dynamics alanine scanning of amyloid-forming peptides in explicit solvent system
Satoshi Kosuda[1], Atsushi Suenaga[2], Gentaro Morimoto[3], Makoto Taiji[3], Yutaka Kuroda[1] ([1]*Dep. Biotech. Life Sci., Grad. Sch. Eng., TUAT.*, [2]*Molprof, AIST*, [3]*QBiC, RIKEN*)

3Pos011 **エステル基を有する新規白金系複核制がん剤による DNA の高次構造と遺伝子活性への影響**
Action of novel anticancer-active, dinuclear platinum complexes with ester group on the higher order structure and genomic activity of DNA
Yuta Shimizu[1], Yuko Yoshikawa[2], Takahiro Tsuchiya[3], Hiroki Yoneyama[4], Shinya Harusawa[4], Seiji Komeda[3], Tadayuki Imanaka[2], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Univ. Doshisha*, [2]*Univ. Ritsumeikan*, [3]*Univ. Suzuka Med. Sci.*, [4]*Univ. Osaka Pharm. Sci.*)

3Pos012 **Attempts at CA-type formal analysis of fibrous assembly of particles**
Takashi Konno (*Univ. Fukui. Med. Mol. Physiol.*)

3Pos013 **The first crystal structure of intact 3.8 MDa molluscan hemocyanin**
Asuka Matsuno[1], Zuoqi Gai[2], Koji Kato[1,2], Sanae Kato[3], Takeshi Shimizu[4], Takeya Yoshioka[4], Hideki Kishimura[5], Tohru Terada[6], Yoshikazu Tanaka[1,2], Min Yao[1,2] ([1]*Grad. Schl. of Life Sci., Hokkaido Univ*, [2]*Facl. of Adv. Life Sci., Hokkaido Univ*, [3]*Asahikawa Med. Univ*, [4]*Hokkaido Ind. Tech. Cent*, [5]*Grad. Schl. of Fish. Sci*, [6]*Grad. Schl. of Agr. and Life Sci., The Univ. of Tokyo*)

3Pos014 **二面角系基準振動解析プログラムの並列化と巨大分子への適用**
Parallelization of the program for normal mode analysis in torsional angle space and application to supramolecules
Shigeru Endo[1], Hiroshi Wako[2] ([1]*Dept. Phys., Sch. Science, Kitasato Univ.*, [2]*Sch. Social Sciences, Waseda Univ.*)

3Pos015 **Template based protein modeling using a target dependent template library**
Kodai Takagi, George Chikenji, Yota Masuyama (*Grad. Sch. Eng., Nagoya Univ.*)

3Pos016 **レプリカ交換アンブレラサンプリング MD シミュレーションを用いたヒストン脱アセチル化酵素阻害剤の選択性の研究**
Study for the selectivity of a histone deacetylase inhibitor using replica-exchange umbrella sampling MD simulation
Shuichiro Tsukamoto[1], Yoshitake Sakae[1], Yukihiro Itoh[2], Takayoshi Suzuki[2,3], Yuko Okamoto[1,3,4,5,6] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch. Med. Sci., Kyoto Pref. Univ. Med.*, [3]*JST-CREST*, [4]*Struc. Biol. Res. Center, Grad. Sch. Sci., Nagoya Univ.*, [5]*Center Comput. Sci., Grad. Sch. Eng., Nagoya Univ.*, [6]*Info. Tech. Center, Nagoya Univ.*)

蛋白質：構造･機能 / Protein: Structure & Function

3Pos017 **Interaction of Two Subunits of D/L Hetero Dimer with Catalytic Subunits in Archaeal RNA polymerase: Insights from MD Simulations**
Neetha Mohan[1], Kota Kasahara[1], Akira Hirata[2], Haruki Nakamura[1] ([1]*Laboratory of Protein Informatics, Institute for Protein Research, Osaka University*, [2]*Department of Materials and Biotechnology, Graduate School of Science and Engineering, Ehime University*)

3Pos018 **光活性化アデニル酸シクラーゼ合成酵素 PAC の活性化機構解明**
Structural and functional insights into a photoactivated adenylyl cyclase
Mio Ohki[1], Kanako Sugiyama[1], Fumihiro Kawai[1], Shigeru Matsunaga[2], Naoya Shibayama[3], Mineo Iseki[4], Sam-Yong Park[1] ([1]*Grad. Sch. of Medi. Life Sci, Yokohama City Univ.*, [2]*Cent. Research Lab. Hamamatsu Photonics K.K.*, [3]*Dept. of Physiology Jichi Medi. Univ.*, [4]*Faculty of Pharma. Sci. Toho Univ.*)

3Pos019 マウス微小ウイルスの力学的性質：粗視化分子動力学シミュレーション
Mechanical properties of the minute mice of virus capsid: Coarse-grained Molecular simulation
Koji Ono, Shoji Takada (*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ*)

3Pos020 Momorcharin の酵素活性を制御する活性部位近傍の特殊なコンフォメーション
The specific conformation near the active site regulating the enzymatic activity of momorcharin
Yuki Okada[1], Ayana Okuno[2], Etsuko Nishimoto[3] ([1]*Grad. Sch. Bioresour. Bioenviron. Sci., Univ. Kyushu*, [2]*Sch. Agr., Univ. Kyushu*, [3]*Fac. Agr., Univ. Kyushu*)

3Pos021 ケモカイン受容体の細胞内領域に含まれるケモカインシグナル制御因子フロント結合領域の構造生物学的解析
Structural analyses of the cytosolic region of the chemokine receptor, which interacts with the chemokine signal regulator FROUNT
Sosuke Yoshinaga[1], Kaori Esaki[1], Tatsuichiro Tsuji[1], Etsuko Toda[2], Yuya Terashima[2], Takashi Saitoh[3], Daisuke Kohda[3], Toshiyuki Kohno[4], Masanori Osawa[5], Takumi Ueda[5], Ichio Shimada[5], Kouji Matsushima[2], Hiroaki Terasawa[1] ([1]*Fac. Life Sci., Kumamoto Univ.*, [2]*Grad. Sch. Med., Univ. Tokyo*, [3]*Med. Inst. Bioreg., Kyushu Univ.*, [4]*Kitasato Univ. Sch. Med.*, [5]*Grad. Sch. Pharm. Sci., Univ. Tokyo*)

3Pos022 酵素と基質と四つのイントロンで形成される平面の関係
Enzyme, Ligand, and Plane formed with Four Introns
Michiko Nosaka, Akari Ichisima, Mami Nakayashiki (*National Institute of Technology, Sasebo College*)

3Pos023 酵母カーゴ様タンパク質受容体 Emp46p と Emp47p のコイルドコイル領域における pH 依存的な会合と解離
pH-dependent assembly and segregation of the coiled-coil segments of yeast putative cargo receptors Emp46p and Emp47p
Kentaro Ishii[1], Hiroki Enda[2], Masanori Noda[3], Megumi Kajino[2], Akemi Kim[2], Eiji Kurimoto[2,4], Ken Sato[5], Akihiko Nakano[6,7], Yuji Kobayashi[3], Hirokazu Yagi[2], Susumu Uchiyama[1,3], Koichi Kato[1,2,8] ([1]*Okazaki Inst. Integrative Bioscience, NINS*, [2]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [3]*Grad. Sch. Eng., Osaka Univ.*, [4]*Fac. Pharm., Meijo Univ.*, [5]*Grad. Sch. Art. Sci., Univ. Tokyo*, [6]*Grad. Sch. Sci., Univ. Tokyo*, [7]*RIKEN, Cent. Phot.*, [8]*Inst. Mol. Sci., NINS.*)

3Pos024 クライオ電子線トモグラフィー法により明らかになったイネ萎縮ウイルスの昆虫細胞からの放出メカニズム
Mechanisms for egress of Rice dwarf virus from insect vector cells revealed by whole cell cryo-electron tomography
Naoyuki Miyazaki[1,2], Akifumi Higashiura[2], Kazuyoshi Murata[1], Atsushi Nakagawa[2], Kenji Iwasaki[2] ([1]*NIPS*, [2]*IPR*)

3Pos025 **Dynamics of bacterial flagellar filament self-assembly**
Anthony Abraham, Takuma Fukumura, Tohru Minamino, Keiichi Namba (*Osaka Uni. Grad. Sch. Frontier Biosciences*)

3Pos026 α シヌクレインの添加物によるアミロイド線維形成への影響
The effects additives on the amyloid formation ofα-synuclein
Miki Hirano[1], Masatomo So[1], Hisashi Yagi[3], Yasushi Kawata[2], Yuji Goto[1] ([1]*Institute for Protein Research, Osaka University*, [2]*Dept. of Chem. and Biotech., Grad. Sch. of Eng., Tottori Univ.*, [3]*Center for Research on Green Sustainable Chemistry, Tottori University*)

3Pos027 １アミノ酸置換によるプロリン異性化酵素からタンパク質分解酵素への機能転換
Functional conversion from peptydyl-prolyl isomerase to protease by a single amino acid substitution
Teikichi Ikura, Nobutoshi Ito (*MRI, Tokyo Med. Dent. Univ.*)

3Pos028 粗視化力場を用いたタンパク質構造変化を伴うリガンド結合過程の比較解析
A comparative study of the protein-ligand binding processes coupled to protein conformational changes in coarse-grained simulations
Tatsuki Negami, Tohru Terada, Kentaro Shimizu (*Grad. Sch. of Agri. and Life Sci., Univ. of Tokyo*)

3Pos029 脂質による GPCR の活性制御機構の解明
Elucidation of the signal regulation mechanism of GPCRs under physiological lipid bilayer environments
Takuya Mizumura[1], Keita Kondo[1], Takumi Ueda[1,2], Yutaka Kofuku[1], Ichio Shimada[1] ([1]*Grad. Sch. Pha., Univ. Tokyo*, [2]*JST-PRESTO*)

3Pos030 滴定 X 線溶液散乱測定を用いたアダプター蛋白質 GGA-ユビキチン複合体の相互作用/構造解析
Interaction/Structure analysis of GGA-Ubiquitin complex by using titration SAXS Measurement
Miho Shinohara[1], Hironari Kamikubo[1], Keito Yoshida[1], Yoichi Yamazaki[1], Kazuhisa Nakayama[2], Soichi Wakatsuki[3], Mikio Kataoka[1] ([1]*Grad. Sch. Mat. Sci., NAIST*, [2]*Grad. Sch. Pharmaceutical Sciences., Kyoto Univ.*, [3]*School of Medicine, Stanford University*)

3Pos031 LigMap: 共溶媒分子シミュレーションによるタンパク質のリガンド結合ホットスポットの予測
LigMap: Predicting ligand binding hostspots of proteins by molecular simulations with cosolvent solutions
Hironori Kokubo, Akihiro Yokota, Atsutoshi Okabe (*Takeda Pharmaceutical*)

3Pos032 貝由来炭酸脱水酵素ナクレインのカルシウム結合部位の解析
Structural Analysis of Calcium Binding Site in Sea Shell Carbonic Anhydrase, Nacrein
Hideto Shimahara[1], Muhamad Koyimatu[1], Yuji Kobayashi[2] ([1]*JAIST CNMT*, [2]*Osaka Univ.*)

3Pos033 **Relative Binding Free Energy Calculation with the Free Energy Variational Principle for fXa-Ligand system**
Toshimichi Matsugi, Takeshi Ashida, Takeshi Kikuchi (*Ritsumeikan Univ*)

3Pos034 高速 AFM による c-Cbl の構造変化の直接観察
Direct observation of conformational change of c-Cbl by high speed AFM
Yoshiki Takahashi[1], Jun-ichi Kishikawa[2], Hiromi Tanaka[3], Hiroki Konno[3] ([1]*Grad. Sch. of Nat. Sci., Kanazawa Univ.*, [2]*Fac. Life Sci., Kyoto Sangyo Univ.*, [3]*Bio-AFM Frontier Research Center, Kanazawa Univ.*)

蛋白質：特性 / Protein: Property

3Pos035 **Motion Tree によるタンパク質運動の階層的記述**
Motion Tree delineates hierarchical structure of protein dynamics observed in molecular dynamics simulation
Kei Moritsugu[1], Ryotaro Koike[2], Akinori Kidera[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City University*, [2]*Grad. Sch. of Info. Sci., Nagoya University*)

3Pos036 **ATP 結合に伴う GroEL 立体構造変化の計算科学的研究**
Computational study of conformational change in GroEL upon ATP binding
Yuka Suzuki[1,2], Kei Yura[3,4,5] ([1]*Dept. Biol., Ochanomizu Univ.*, [2]*OIST*, [3]*Grad. School of Humanities and Sciences, Ochanomizu Univ.*, [4]*Center for Info. Biol., Ochanomizu Univ.*, [5]*NIG*)

3Pos037 **分子動力学法による好熱性ロドプシンの熱安定性の研究**
Assessing Molecular Mechanism of High Thermal Stability of Thermophilic Rhodopsin by Molecular Dynamics Simulation
Taisuke Hasegawa[1], Yuki Sudo[2], Takeshi Murata[3], Shigehiko Hayashi[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*, [3]*Grad. Sch. Sci., Chiba Univ.*)

3Pos038 **Dissociation and Unfolding of Tobacco Mosaic Virus Coat Protein Assemblages**
Hiroaki Fukao[1], Kazumasa Sakurai[2], Yasushige Yonezawa[2], Masao Fujisawa[1], Kazuhiro Ishibashi[3], Masayuki Ishikawa[3], Tetsuo Meshi[3], Hideki Tachibana[1,2] ([1]*Fac Boil-Ortd Sci Tech, Kinki Univ*, [2]*High-Pres Prot Res Center, Kinki Univ*, [3]*Div Plant Sci, NIAS*)

3Pos039 **NMR で見た水溶液中メリチンの１量体ー4 量体構造転移**
Conformational transition between monomer and tetramer of melittin in an aqueous solution studied by NMR
Yoshinori Miura (*Center for Advanced Instrumental Analysis, Kyushu University*)

3Pos040 **酸ストレス抗体の中性および穏和な温度における凝集の速度論**
Kinetics of antibody aggregation triggered by pH-shift stress at neutral pH and ambient temperatures
Hiroshi Imamura, Shinya Honda (*National Institute of Advanced Industrial Science and Technology*)

3Pos041 **Role of electrostatic repulsion between unique arginine residues on the assembly of a trimeric autotransporter translocator domain**
Eriko Aoki, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. Bioinfo., Grad. Sch. Eng., Soka Univ.*)

3Pos042 **熱測定を用いたグルカゴン凝集反応の熱力学的研究**
Thermodynamic characterization of glucagon aggregation using calorimetry
Tetsuhei Uenoyama[1], Tatsuya Ikenoue[1], Daniel E Otzen[2], Yuji Goto[1], Young-Ho Lee[1] ([1]*Inst. for. Prot. Research, Osaka Univ*, [2]*Department of Molecular Biology, Aarhus Univ*)

3Pos043 **Molecular Tailoring Approach 法による蛋白質二次構造内に働く相互作用の量子化学的研究**
Quantum chemical study for interactions in protein secondary structures via Molecular Tailoring Approach
Ayumi Kusaka[1,2], Haruki Nakamura[1], Yu Takano[1,3,4] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Grad. Sch. Info. Sci., Hiroshima City Univ.*, [4]*JST-CREST*)

3Pos044 **神経細胞毒性を持つ $A\beta_{42}$ 凝集体と持たない凝集体は共に β 構造から成るが短波長領域で違った CD パターンを持っている。**
Neurotoxic and nontoxic $A\beta_{42}$ assemblies are commonly composed of β-sheet structures but have different CD spectra in the short wavelength
Yoshitaka Matsumura[1,2], Tomoya Sasahara[1,2], Takayuki Ohnishi[1,2], Eri Saijo[1,2], Kaori Satomura[1,2], Michio Sato[3], Minako Hoshi[1,2,4] ([1]*TAO Health Life Pharma Co., Ltd.*, [2]*Inst. Biomed. Research & Innov.*, [3]*Meiji Univ.*, [4]*Kyoto Univ.*)

3Pos045 **レプリカ交換分子動力学シミュレーションによる ポリグルタミン酸の pH に対する構造依存性**
Structural dependence of poly-glutamic acids on pH studied by replica-exchange molecular dynamics simulations
Ryosuke Iwai[1], Tetsuro Nagai[2], Takuya Takahashi[2] ([1]*Grad. Sci. Life Sci., Ritsumeikan Univ*, [2]*Coll. Life. Sci., Ritsumeikan Univ*)

蛋白質：機能 / Protein: Function

3Pos046 **枯草菌 F_1-ATPase に於ける DELSEED 領域の機能解析**
Role of the DELSEED motif in *Bacillus subtilis* F_1-ATPase
Koji Takada, Yasuyuki Kato-Yamada (*Department of Life Science, Rikkyo University*)

3Pos047 **MD シミュレーションを用いた Neuropsin へのペプチド結合に関する研究**
Molecular Dynamics Study of Peptide Binding to Neuropsin
Masami Lintuluoto[1], Mitsumasa Abe[1], Hideki Tamura[2], Yoshifumi Fukunishi[3] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ*, [2]*L-StaR, Hoshi Univ. Sch. Pham. and Pham. Sci.*, [3]*AIST*)

3Pos048 **アクチン結合タンパク質アクチニン-４とその変異体の機能解析**
Predicting the three-dimensional structure of the actin-binding domains of actinin-4 mutants
Nami Miura, Masahiro Kamita, Takanori Kakuya, Hirokazu Shoji, Tesshi Yamada, Kazufumi Honda (*Div. Chem. Clin., Natl. Cancer Ctr. Res. Inst.*)

3Pos049 **MD シミュレーションを用いた G3LEA モデルペプチドとタンパク質の相互作用の解析**
Analysis of Group3LEA model peptide-protein interactions by molecular dynamics simulation
Makoto Usui, Takao Furuki, Tadaomi Furuta, Minoru Sakurai (*Center for Biological Resources and Informatics, Tokyo Institute of Technology*)

3Pos050 **F1-ATPase における ε サブユニットの活性制御因子としての役割**
Role of ε subunit as a regulator for F1-ATPase
Makoto Genda[1], Rikiya Watanabe[1], Yasuyuki Yamada[2], Hiroyuki Noji[1] ([1]*Grad. Sch. Eng. , Univ. Tokyo*, [2]*Dept. Sci. , Univ. Rikkyo*)

3Pos051 **アデニル酸キナーゼ反応機構に関する計算科学的研究**
Computational Study on the Reaction Mechanism of Adenylate Kinase
Kenshu Kamiya (*Dept. Phys., Sch. Sci., Kitasato Univ.*)

蛋白質：計測･解析 / Protein: Measurement & Analysis

3Pos052 **高精度で高効率な水和自由エネルギー計算法**
An accurate and efficient method to compute the hydration free energy for large and complex molecules
Takashi Yoshidome[1], **Toru Ekimoto**[1], Nobuyuki Matubayasi[2], Yuichi Harano[3], Masahiro Kinoshita[4], Mitsunori Ikeguchi[1] ([1]*Yokohama City Univ.*, [2]*Osaka Univ.*, [3]*Himeji Dokkyo Univ.*, [4]*Kyoto Univ.*)

3Pos053 **分子シミュレーションにおける静電相互作用計算のための新規非エバルト法**
Novel non-Ewald methods for calculating electrostatic interactions in molecular simulations
Ikuo Fukuda[1], Narutoshi Kamiya[1], Han Wang[2], Kota Kasahara[1], Haruki Nakamura[1] ([1]*Institute for Protein Research, Osaka University*, [2]*Freie Universitaet Berlin*)

3Pos054 **再構築型無細胞タンパク質合成系を用いた 真核生物由来タンパク質の凝集性の大規模解析**
Large-scale analysis of aggregation propensities of eukaryotic proteins by using a reconstituted cell-free translation system
Tatsuya Niwa[1], Eri Uemura[1], Kazuhiro Takemoto[2], Shintaro Minami[3], Satoshi Fukuchi[4], Motonori Ota[3], Takuya Ueda[5], Hideki Taguchi[1] ([1]*Grad. Sch. Biosci. and Biotech., Tokyo Tech*, [2]*Dept. Biosci. and Bioinfo., Kyushu Institute of Technology*, [3]*Grad. Sch. Inf. Sci., Nagoya Univ.*, [4]*Faculty Eng., Maebashi IT*, [5]*Grad. Sch. Frontier Sci., Univ. Tokyo*)

3Pos055 **複数の正則化項を用いた圧縮センシングによる NMR スペクトルの再構成**
Reconstruction of NMR spectra using compressed sensing with multiple regularization terms
Kazuya Sumikoshi[1], Teppei Ikeya[2], Yutaka Ito[2], Kentaro Shimizu[1] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci. Eng., Tokyo Metropolitan Univ.*)

3Pos056 **アミロイドβ凝集体に結合したチオフラビン T の蛍光寿命**
Fluorescence lifetime of Thioflavin T binding to amyloid-beta peptide aggregates
Akinori Oda, Hiroshi Satozono, Tomomi Shinke, Yohei Takata, Hiroyuki Okada (*Hamamatsu Photonics K.K.*)

3Pos057 **固体 NMR を用いたユビキチン過剰発現時に単一大腸菌細胞内で合成される分子数の計測**
Counting of the molecules synthesized in an ubiquitin-overexpressed intact *Escherichia coli* cell by solid-state NMR
Kazuya Yamada, Ayako Egawa, Toshimichi Fujiwara (*IPR, Osaka Univ.*)

蛋白質：工学 / Protein: Engineering

3Pos058 **抗酸化亜鉛 VHH 抗体の亜鉛イオン結合特性**
Zinc ion binding activity of an anti-ZnO VHH antibody, 4F2
Ryosuke Sasaki[1], Soichiro Kitazawa[2], Ryo Kitahara[2], Yoshikazu Tanaka[3], Izumi Kumagai[4], Mitsuo Umetsu[4], **Koki Makabe**[1] ([1]*Yamagata Univ.*, [2]*Ritsumeikan Univ.*, [3]*Hokkaido Univ.*, [4]*Tohoku Univ.*)

3Pos059 **テトラヒメナビオチン化タンパク質の同定と外腕ダイニン運動系への適用**
Identification of biotin carboxyl carrier protein in Tetrahymena and its application in in vitro motility systems of outer arm dynein
Masaki Edamatsu (*Grad. Sch. Arts Sci. Univ. Tokyo*)

3Pos060 **細胞透過性コイルドコイルタンパク質の熱安定性**
Thermal stability of coiled-coil cell-penetrating proteins
Tsubasa Yuki[1], Norihisa Nakayama[1], **Ken-Ichi Sano**[1,2] ([1]*Grad. Sch. Env. Sys., Nippon Inst. Tech.*, [2]*Dept. Innovative Sys. Eng. Nippon Inst. Tech.*)

3Pos061 **Selection of RuBpy3 motifes from a randomized peptide library**
Marziyeh Karimiavargani[1], Noriko Minagawa[2], Takuji Hirose[1], Yoshihiro Ito[2], Takanori Uzawa[2] ([1]*Univ. Saitama*, [2]*Nanomedical. Riken*)

ヘム蛋白 / Heme proteins

3Pos062 **ヒト cytochromeb561 ホモログタンパク質・ヒト SDR2 の生理機能解析**
Functional analysis of human stromal cell-derived receptor 2, a homolog of cytochrome b561
Rei Toda, Yuma Takahashi, Takako Yamazoe, Akikazu Asada, Motonari Tsubaki (*Dept. of Chem., Grad. Sch. of Sci., Kobe Univ.*)

3Pos063 **QM/MM と MD シミュレーションを用いた黄色ブドウ球菌の細胞壁上の高速ヘム輸送機構の解明**
Rapid heme-transfer reactions across the cell wall of *Staphylococcus aureus*: a theoretical study using QM/MM and MD simulations
Yoshitaka Moriwaki, Tohru Terada, Kentaro Shimizu (*Dept. Biotech., Grad. Sch. Agri., Univ. of Tokyo*)

3Pos064 **共鳴ラマン分光法による 2 価コバラミンの pH に依存した構造変化の検出**
Resonance Raman Detection of Cobalamin (II) : pH-Dependent Structural Change
Kaoru Mieda, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

3Pos065 **呼吸鎖 O_2 還元酵素反応中間体の構造解析**
Structural analysis of respiratory O_2 reductase in the reaction intermediate state
Kazumasa Muramoto (*Dept. of Life Sci., Univ. of Hyogo*)

膜蛋白質 / Membrane proteins

3Pos066 **光照射固体 NMR による 13-cis, 15-syn フォトサイクル及び中間体の解明**
Elucidation of 13-cis, 15-syn photocycle and its intermediate by photo-irradiation ss-NMR
Arisu Shigeta[1], Kyosuke Oshima[1], Izuru Kawamura[1], Takashi Okitsu[2], Akimori Wada[2], Satoru Tuzi[3], Akira Naito[1] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Univ. Hyogo*)

3Pos067 **G 蛋白質共役受容体を熱安定化させるアミノ酸置換の予測：自由エネルギー関数の開発**
Prediction of Thermostabilizing Mutations for G Protein-Coupled Receptors: Development of Free-energy Function
Satoshi Yasuda[1], Yuta Kajiwara[2], Yuuki Takamuku[3], Nanao Suzuki[3], Takeshi Murata[3], Masahiro Kinoshita[1] ([1]*Institute of Advanced Energy, Kyoto Univ.*, [2]*Graduate School of Energy Science, Kyoto Univ.*, [3]*Graduate School of Science, Chiba Univ.*)

3Pos068 **アクアポリン 1 の細孔口における水分子のダイナミクス**
Water dynamics at channel entrance of aquaporin 1
Eiji Yamamoto[1], Takuma Akimoto[1], Masato Yasui[2], Kenji Yasuoka[3] ([1]*Department of Mechanical Engineering, Keio University*, [2]*School of Medicine, Keio University*, [3]*Department of Mechanical Engineering, Keio University*)

3Pos069 **FSEC を用いた膜タンパク質複合体 MotA/B の構造安定性評価**
Evaluation of structural stability of membrane protein complex MotA/B by FSEC
Hiroko Takazaki[1], Keiichi Namba[1,2], Takashi Fujii[1,3] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*QBiC RIKEN*, [3]*JST PRESTO*)

3Pos070 **Toward simultaneous observation of conformational dynamics and proton transport in FoF1-ATP synthase**
Naoki Soga[1], Rikiya Watanabe[1,2,3], Mayu Hara[1], Hiroyuki Noji[1,3] ([1]*Dept. of app. chem., The Univ. of Tokyo*, [2]*PRESTO, JST*, [3]*CREST, JST*)

3Pos071 **パッチクランプ法による大腸菌呼吸鎖の末端酸化酵素の H^+-pump の機能解析**
Patch clamp studies on the terminal oxidase of *E. coli*
Masaaki Uno[1], Kosuke Komazawa[1], Teruo Kuroda[2], Hisashi Kawasaki[1], Isamu Yabe[1] ([1]*Grad. School of Adv. Sci. and Tech., Mate. and Life Sci., Tokyo Denki Univ.*, [2]*Grad. School of Med., Dent and Phar Sci., Okayama Univ.*)

3Pos072 **標的分子に適したナノポアの再構成と一分子検出への挑戦**
Reconstitution of a target-matching nanopore and challenge for a single molecule analysis
Hirokazu Watanabe, Ryuji Kawano (*Dep. of Biotec. and Life Sci., Tokyo Univ. of Agr. and Tech.*)

3Pos073 **カロテノイド結合に伴うハロロドプシンの熱安定化**
Thermal stabilization of halorhodopsin by binding of carotenoid
Takanori Sasaki[1], Kaede Suzuki[1], Takashi Kikukawa[2], Makoto Demura[2] ([1]*Sch. Sci. and Tech., Meiji Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*)

3Pos074 **高速原子間力顕微鏡によるバクテリオロドプシン球殻構造体の分子構造の観察**
Molecular arrangement in bacteriorhodopsin vesicles observed by high-speed atomic force microscopy
Yuto Noda, Daisuke Yamamoto (*Grad. Sch. Sci., Fukuoka Univ.*)

核酸結合蛋白質 / Nucleic acid binding proteins

3Pos075 **分子シミュレーションによる転写因子の DNA 上障害物の迂回機構の研究**
How transcription factor bypasses obstacles bound on DNA studied by molecular simulations
Mami Saito[1], Tuyoshi Terakawea[2], Shoji Takada[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*Univ. Columbia*)

3Pos076 **H3 ヒストンテールのアセチル化はその立体構造にどのような影響を与えるか**
How does an acetylation affect the conformation of H3 histone tail?
Jinzen Ikebe[1], Shun Sakuraba[2], Hidetoshi Kono[1] ([1]*MMS, JAEA*, [2]*GSFS, U Tokyo*)

3Pos077 **バイモーダルな mRNA コピー数分布の分子起源**
Molecular origins of bimodal mRNA copy-number distribution
Keisuke Fujita[1], Mitsuhiro Iwaki[1,2], Toshio Yanagida[1,2] ([1]*QBiC, Riken*, [2]*Grad. Sch. of Front. Biosci., Osaka Univ.*)

核酸：構造･特性 / Nucleic acid: Structure & Property

3Pos078 **親水性ポリマーがある混雑効果によって引き起こされた DNA の折り畳み転移: PEG の分子サイズの効果**
Folding transition of DNA induced by crowding effect with hydrophilic polymers: Effect of molecular size of PEG
Shogo Ogata[1], Kenichi Yoshikawa[1], Takahiro Kenmotsu[1], Yuko Yoshikawa[2] ([1]*Doshisha University*, [2]*Ritsumeikan University*)

3Pos079 **Sequence dependence of the stability of single-strand base-stacking**
Ryosuke Murai[1], Hiroaki Hata[2], Akira Suyama[1] ([1]*Grad. Sch. Arts and Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Hyogo*)

3Pos080 **Promotion & Inhibition of Gene-Expression with Polyamines**
Ai Kanemura[1], Yuta Shimizu[1], Yuko Yoshikawa[2], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Doshisha University*, [2]*Ritsumeikan University*)

3Pos081 **Protect Effects of Ascorbic Acid against Double-strand Breaks in Giant DNA Molecules: Comparison among the Damages**
Yue Ma[1], Naoki Ogawa[1], Yuko Yoshikawa[2], Toshiaki Mori[3], Tadayuki Imanaka[2], Kenichi Yoshikawa[1] ([1]*Doshisha University*, [2]*Ritsumeikan University*, [3]*Osaka Prefecture University*)

核酸：相互作用･複合体形成 / Nucleic acid: Interaction & Complex formation

3Pos082 **複製開始タンパク質 DnaA と二本鎖 DNA 相互作用の分子動力学計算による解析**
Interaction of replication initiator protein DnaA with dsDNA studied by molecular dynamics simulation
Masahiro Shimizu, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

3Pos083 **SEVENS 法：ヌクレオソームの局所密度の違いでクロマチンを分画する**
SEVENS assay: a chromatin fractionation based on the local density of nucleosomes
Satoru Ishihara (*Fujita Health Univ. Sch. Med.*)

3Pos084 **酵素を用いた DNA 反応系によりつくられた XOR 演算回路**
XOR circuit constructed using enzymatic DNA reactions
Toshihiro Kojima, Yoko Sakai, Koh-ichiroh Shohda, Akira Suyama (*Univ. of Tokyo*)

水･水和･電解質 / Water & Hydration & Electrolyte

3Pos085 **分子シミュレーションを用いた２成分溶液系における選択的溶媒和の部分モルエンタルピーへの影響の研究**
Effects of Preferential Solvation on Partial Molar Enthalpies in Binary Mixture Systems: Molecular Simulation Study
Yuichi Kawabata, Ryo Akiyama (*Grad. Sch. Sci., Univ. Kyushu*)

3Pos086 **The first application of adaptive quantum mechanics / molecular mechanics method for infrared spectra of liquid phase**
Hiroshi Watanabe[1], Misa Banno[1], Kubar Tomas[2], Elstner Marcus[2], Minoru Sakurai[1] ([1]*Tokyo Inst. of Tech.*, [2]*Karlsruhe Inst. of Tech.*)

3Pos087 **3 次元分布相関を基にしたタンパク質水和構造予測法の開発**
Development of structural sampling for predicting hydration structure around the protein, based on 3-dimensional distribution function
Shunsuke Chiba, **Yasuomi Kiyota**, Mayuko Takeda-Shitaka (*Kitasato University*)

3Pos088 **溶質分子周囲の水分子ダイナミクスの MD シミュレーション：人工的に導入された LJ ポテンシャルパラメタ変化の影響**
MD simulations of water dynamics around solute molecules: Effect of LJ potential parameter changes artificially introduced
Takuya Takahashi, Tetsuro Nagai (*Coll. Life. Sci., Ritsumeikan Univ.*)

発生･分化 / Development & Differentiation

3Pos089 **Wnt/β-catenin と FGF/ERK シグナルは再生皮膚の羽毛原基の発生とパターニングに関与する**
Wnt/β-catenin and FGF/ERK signaling are involved in the feather bud formation and patterning of reconstructed skin
Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

3Pos090 **Mapping elastic modulus of mouse submandibular gland tissue by atomic force microscopy : effect of cytoskeletal modification**
Yu Nakajima[1], Mitsuhiro Nakamura[1], Hiroaki Taketa[2], Takuya Matsumoto[2], Takaharu Okajima[1] ([1]*Grad. Sch. of Inf and Sci., The Univ. of Hokkaido*, [2]*Grad. Sch. Med. Den. and Pham. Sci.,Univ. Okayama*)

3Pos091 **C. elegans 受精卵の極性維持には RING タンパク PAR-2 の膜-細胞質間の local な交換反応が機能する**
Cortical polarity gradient maintenance by local-rapid cortex-cytoplasm exchange of a posterior PAR
Yukinobu Arata[1], Michio Hiroshima[1,2], Chan-Gi Pack[1,8], Ravikrishna Ramanujam[3], Fumio Motegi[3], Kenichi Nakazato[4], Hitoshi Sawa[5], Tetsuya J. Kobayashi[6], Tatsuo Shibata[7], Yasushi Sako[1] ([1]*Cellular Informatics Laboratory, RIKEN*, [2]*Cell Signaling Dynamics, QBiC, RIKEN*, [3]*Temasek Lifescience Laboratory, Mechanobiology Institute, National University of Singapore*, [4]*Theoretical Biology Laboratory, RIKEN*, [5]*Multicellular Organization, National Institute of Genetics*, [6]*Quantitative biology, Institute of Industrial Science, University of Tokyo*, [7]*Physical Biology, QBiC, RIKEN*, [8]*Asan Institute for Life Sciences, University of Ulsan, College of Medicine, Asan Medical Center*)

筋肉 / Muscle

3Pos092 **横紋筋筋原線維束上の２次元 SPOC 波に関するモデルシミュレーション**
Model simulation on the two dimensional SPOC wave in a bundle of striated myofibrils
Koutaro Nakagome[1], Katsuhiko Sato[2], Shin'ichi Ishiwata[1,3] ([1]*Faculty of Sci and Eng, Waseda Univ*, [2]*RIES, Hokkaido Univ*, [3]*WABIOS*)

3Pos093 **水晶振動子上でのアクトミオシンの質量**
Weighing the apparent mass of actomyosin by QCM
Kazuya Soda[1], Takashi Ishiguro[2], Hajime Honda[1] ([1]*Dept. of Bioeng., Nagaoka Univ. Tech.*, [2]*Taiyo Yuden Co., Ltd.*)

3Pos094 **ミオシンサブフラグメント 1 の水和に及ぼす ATP アナログの影響**
Hydration analysis on myosin subfragment-1 with ATP-analogs
Hideyuki Ohsugi[1], George Mogami[1], Tetsuichi Wazawa[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Inst. Sci. Ind. Res., Osaka Univ.*)

3Pos095 **Mg ポリマー再考２**
Revisiting "Mg-Polymer" 2
Mahito Kikumoto, Shuichi Takeda, Yuiticho Maeda (*Struct. Biol. Center, Grad. Sch. Sci., Nagoya-U.*)

3Pos096 **アクチンの集団運動内でのアクチン間の長距離相互作用**
Long range interaction among of actin filaments under their collective movement
Yuto Fujita, Shigeru Sakurazawa (*Grad. Sch. System Info., Future Univ. Hakodate*)

分子モーター / Molecular motor

3Pos097 **Inter-subunit coordination around a ring-shaped ATPase**
Liqiang Dai, Jin Yu (*Beijing Computational Science Research Center*)

3Pos098 **Dynamic instability of microtubules in a ROS free environment**
Md. Sirajul Islam[1], Arif Md. Rashedul Kabir[2], Daisuke Inoue[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

3Pos099 **DNA programmed active self-organization of microtubules on kinesin**
Jakia Jannat Keya[1], Daisuke Inoue[2], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Faculty of Sci., Hokkaido Univ.*)

3Pos100 **Observation of the gliding machinery of Mycoplasma mobile by Quick-Freeze Deep-Etch Replica Electron Microscopy**
Clothilde Bertin, Yuhei O. Tahara, Tasuku Hamaguchi, Eisaku Katayama, Makoto Miyata (*Grad. School Sci., Osaka City Univ.*)

3Pos101 **V1-ATPase の A3B3 固定子における鞭毛タンパク質 FliJ の回転軸機能**
Rotor function of flagella protein FliJ in A3B3 of V1-ATPase
Mihori Baba[1], Atsuko Nakanishi[1], Jun-ichi Kishikawa[1], Shou Furuike[2], Ken Yokoyama[1] ([1]*Dept. Mol. Biosci., Kyoto Sangyo Univ.*, [2]*Dept. Phys, Osaka Medical College*)

3Pos102 **試験管内再構築系を用いた細胞膜-アクチンコーテックス複合体変形機構の解明**
Reconstituted actomyosin cortex deformed in size dependent manner
Yukinori Nishigami[1], Hiroaki Ito[1], Seiji Sonobe[2], Masatoshi Ichikawa[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Univ. Hyogo*)

3Pos103 **高速原子間力顕微鏡を用いた回転軸の無い V_1-ATPase のコンフォメーション変化の観察**
Observation of Conformational changes of rotorless V_1-ATPase using High-Speed Atomic Force Microscopy
Motonori Imamura[1], Kazuya Nakamoto[2], Takayuki Uchihashi[1,3,4], Takeshi Murata[2,5], Toshio Ando[1,3,4] ([1]*Dept. of Phys., Kanazawa Univ.*, [2]*Grad. Sch. Sci., Chiba Univ.*, [3]*Bio-AFM FRC, Kanazawa Univ.*, [4]*CREST, JST*, [5]*PRESTO, JST*)

3Pos104 **アクチンフィラメントに対する HMM とコフィリンの相互排他的結合の定量的解析**
A quantitative analysis of exclusive binding of HMM and Cofilin to actin filament
Hiroaki Ueno[1], Yuusuke Nishikawa[1], Rika Hirakawa[1], Taiga Imai[1], Taro Q.P. Uyeda[2], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Sustain. Environ. Eng., Muroran Inst.*, [2]*Biomedical Res. Inst., AIST*)

3Pos105 **フォトクロミック分子で変化する有糸分裂キネシン Eg5 の ATP 分解活性経路の光制御ステップのストップドフロウ解析**
Stopped flow analysis on the photo-regulated step in ATPase kinetic pathway of mitotic kinesin Eg5 modified with photochromic molecule
Kentaro Saito, Kei Sadakane, Yuki Tamura, Ryoma Yamamoto, Shinsaku Maruta (*Grad. Sch. Eng., SOKA Univ.*)

3Pos106 **キネシン Eg5 の新規フォトクロミック阻害剤存在下における運動アッセイと速度論的解析**
Kinetic analysis on the mitotic kinesin Eg5 ATPase and in vitro motility assay in the presence of a novel photochromic inhibitor
Kei Sadakane, Yuhki Tamura, Kentaro Saitoh, Ryoma Yamamoto, Shinsaku Maruta (*Soka University, Graduate school of Engineering, Division of Bioinformatics*)

3Pos107 **好熱菌由来の回転モーター F_1 のカップリングスキーム**
Coupling scheme of the rotary motor thermophilic F_1
Kengo Adachi[1], Kazuhiro Oiwa[2], Masasuke Yoshida[3], Kazuhiko Kinosita, Jr.[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Adv. ICT Res. Inst., NICT*, [3]*Dep. Mol. Biosci., Kyoto Sangyo Univ.*)

3Pos108 **The structure of the flagellar filament of magnetotactic bacterium MO-1 by electron cryomicroscopy**
Juanfang Ruan[1], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*QBiC, RIKEN*)

3Pos109 **鞭毛内腕ダイニンの形成にはアクチンの N 末端側が重要である**
N-terminal sequence of actin is critical for the assembly of flagellar inner-arm dyneins
Takako Kato-Minoura, Yuko Horikoshi, Kaori Imaeda (*Dept. Biol. Sci., Chuo Univ.*)

3Pos110 **Single-molecule analysis of hybrid *Enterococcus hirae* V_1-ATPase toward elucidation of the chemo-mechanical coupling scheme**
Yoshihiro Minagawa[1], Hiroshi Ueno[1], Hiroyuki Noji[1], Takeshi Murata[2], Ryota Iino[3,4] ([1]*Dept. App. Chem., Grad. Sch. Eng., The Univ. Tokyo*, [2]*Grad. Sch., Univ. Chiba*, [3]*Okazaki Inst. Integ. Biosci., NINS*, [4]*Grad. Univ. for Adv. Studies,*)

3Pos111 **マイコプラズマ滑走タンパク質 Gli349 を構成するリピート断片の立体構造解析**
Structural analysis of repeat fragments consisting of the gliding protein Gli349 from *Mycoplasma mobile*
Junichi Inatomi[1], **Yuuki Hayashi**[1], Yoshihiro Nomura[2], Yoshito Kawakita[3], Masaru Yabe[3], Masato Miyata[3], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Integrated Sci., Univ. Tokyo*, [3]*Dept. Biol., Osaka City Univ.*)

3Pos112 **腸内連鎖球菌由来 V-ATPase の Na^+濃度依存の回転**
Na^+-dependent rotation of *Enterococcus hirae* V-ATPase
Hiroshi Ueno[1], Yoshihiro Minagawa[1], Hiroyuki Noji[1], Takeshi Murata[2], Ryota Iino[3,4] ([1]*Dept. App. Chem.,Grad. Sch. Eng., The Univ. Tokyo*, [2]*Dept. Chem., Grad. Sch. Sci., Univ. Chiba*, [3]*Okazaki Inst. Integ. Biosci., IMS, NINS*, [4]*Dept. of Functional Molecular Science, SOKENDAI*)

3Pos113 **カーボンナノチューブを用いたべん毛モーターの抑制的制御**
Carbon nanotube-based deactivation of bacterial flagellar motors
Yuichi Inoue[1], Yoichiro Sawano[2], Hajime Fukuoka[3], Hiroto Takahashi[1], Ishijima Akihiko[1,3] ([1]*IMRAM, Tohoku Univ*, [2]*Grad. Sch. Life Sci., Tohoku Univ.*, [3]*Grad. Sch. Front. Biosci, Osaka Univ.*)

3Pos114 **ゆらぎ計測によるゼブラフィッシュ色素顆粒輸送の力-速度関係**
Force-velocity relation of organelle transport in Zebrafish melanophores: new fluctuation analysis
Shin Hasegawa[1], Kazuho Ikeda[2], Yasushi Okada[2], Kumiko Hayashi[1] ([1]*Sch. Eng., Tohoku Univ.*, [2]*QBiC, RIKEN*)

3Pos115 **ダイニンによって駆動される微小管の polar あるいは nematic 集団運動**
Polar or nematic motion of collective microtubules driven by dyneins
Naoki Kanatani[1], Takayuki Torisawa[2,3], Hiroaki Kozima[2], Kazuhiro Oiwa[1,2,3] ([1]*Univ. Hyogo*, [2]*NICT*, [3]*CREST, Biodynamics*)

3Pos116 **光応答性 DNA を用いた微小管集団運動の時空間制御**
Spatiotemporally controlled collective motion of self-propelled microtubules by using photoresponsive DNA
Ryuhei Suzuki[1], Kyohei Uenishi[1], Daisuke Inoue[2], Kazuki Sada[1,2], Akinori Kuzuya[3], Hiroyuki Asanuma[4], Akira Kakugo[1,2] ([1]*Grad. Sch. Chem. Sci. Eng., Hokkaido Univ.*, [2]*Fac. Sci., Hokkaido Univ.*, [3]*Fac. Chem. Mater. Bioeng., Kansai Univ.*, [4]*Grad. Sch. Eng., Nagoya Univ.*)

3Pos117 **Formation and rupture of a motorized cytoskeletal network**
Daisuke Taniguchi[1,2], Takayuki Torisawa[2,3], Kazuhiro Oiwa[2,3], Shuji Ishihara[1,2] ([1]*Dep. of Physics, Sch. of Science and Technology, Meiji Uni.*, [2]*CREST, JST*, [3]*Advanced ICT Research Institute, NICT*)

3Pos118 **SH1 ヘリックス変異ミオシン II の運動活性と熱安定性**
Motile activity and thermal stability of SH1 helix mutant myosin II
Kotomi Shibata[1], Sosuke Iwai[2], Shigeru Chaen[1] ([1]*Grad. Sch. Sci., Univ. Nihon*, [2]*Edu., Univ. Hirosaki*)

3Pos119 **高速 AFM によるミオシン VI のモーターメカニズムの解明**
Motor mechanism of myosin VI studied by high-speed AFM
Shiori Sano[1], Noriyuki Kodera[2,3], Daniel Safer[4], H.Lee Sweeney[4], Toshio Ando[1,2,5] ([1]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ*, [2]*Bio-AFM FRC, Inst. of Sci. and Eng., Kanazawa Univ*, [3]*PRESTO, JST*, [4]*Dept. of Physiol., Univ. of Pennsylvania Sch. of Med*, [5]*CREST, JST*)

細胞生物学 / Cell biology

3Pos120 **Detection of TRPC and Orai1 proteins in bovine ciliary muscle cells prepared by a Percoll density-gradient centrifugation method**
Motoi Miyazu, Kosuke Takeya, Toshiyuki Kaneko, Akira Takai (*Asahikawa Medical Univ.*)

3Pos121 **ストレス線維における張力依存的な ERK の活性化**
Tension-dependent ERK activation on actin stress fibers
Hiroaki Hirata[1,2], Mukund Gupta[2], Sri Ram Krishna Vedula[2], Chwee Teck Lim[2,3], Benoit Ladoux[2,4], Masahiro Sokabe[2,5] ([1]*R-Pharm Japan*, [2]*Mechanobiol. Inst., Natl. Univ. Singapore*, [3]*Dep. Biomed. Eng., Natl. Univ. Singapore*, [4]*Univ. Paris Diderot*, [5]*Grad. Sch. Med., Nagoya Univ.*)

3Pos122 **表層ストレス応答を制御する膜内切断プロテアーゼ RseP による基質切断反応の生細胞内イメージング**
Live-cell imaging of a proteolytic event by an intramembrane protease RseP that regulates extracytoplasmic stress response
Yohei Hizukuri, Yoshinori Akiyama (*Inst. Virus Res., Kyoto Univ.*)

3Pos123 **ハイドロゲルの表面力学場及び表面生化学の両条件に対する分散培養 iPS 細胞の増殖応答性**
Proliferation response of dissociated iPS cells to the dual parameters of surface mechanics and biochemistry of culture hydrogel
Kenta Mizumoto[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Univ. Kyushu*, [2]*IMCE., Univ. Kyushu*)

3Pos124 **Rotational diffusion of proteins in crowded environment: NMR spectroscopy and molecular dynamics simulation**
Po-hung Wang[1], Isseki Yu[1], Hideyasu Okamura[2], Takanori Kigawa[2], Yuji Sugita[1,3,4,5] ([1]*RIKEN Theoretical Molecular Science Laboratory*, [2]*RIKEN Yokohama*, [3]*RIKEN AICS*, [4]*RIKEN QBiC*, [5]*RIKEN iTHES*)

3Pos125 **フィラミンのアクチン結合ドメインは、極性をもった粘菌細胞後部のアクチンフィラメントを認識し特異的に結合する**
Actin binding domain of filamin recognizes and specifically binds to posterior actin filaments in polarized *Dictyostelium* cells
Keitaro Shibata, **Taro Uyeda** (*Biomed. Res. Inst., AIST*)

3Pos126 **心室・心房の組織片による自律拍動の同期化**
Synchronization of spontaneous beating of tissue fragments of atrium and ventricle
Ryuichi Shinozaki, Tomonori Takahashi, Yuichi Asanuma, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

3Pos127 **細胞サイズ球状閉鎖空間内でのアクトミオシンリングの自発形成と収縮**
Cell-sized spherical confinement induces the spontaneous formation of contractile actomyosin rings *in vitro*
Makito Miyazaki[1], Masataka Chiba[1], Hiroki Eguchi[1], Takashi Ohki[1], Shin'ichi Ishiwata[1,2] ([1]*Dept. of Physics, Waseda Univ.*, [2]*WABIOS, Waseda Univ.*)

3Pos128 **大腸菌走化性シグナル伝達タンパク質の極局在と細胞内シグナル伝達の関係**
Relationship between polar localization of chemotactic proteins and intracellular signaling under steady-state of *Escherichia coli*
Yong-Suk Che[1], Hajime Fukuoka[1], Yuichi Inoue[2], Hiroto Takahashi[2], Akihiko Ishijima[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ*, [2]*IMRAM, Tohoku Univ*)

3Pos129 **バクテリア FlgN シャペロンの機能構造スイッチの分子基盤**
Molecular basis for a structural switch of FlgN that regulates its chaperone activity
Miki Kinoshita[1,2], Yuki Nakanishi[2], Yukio Furukawa[1], Katsumi Imada[2], Keiichi Namba[1,3], Tohru Minamino[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ*, [3]*QBiC, RIKEN*)

3Pos130 **アクチン結合タンパク質によって変化するアクチン繊維内モノマーの位置ゆらぎ**
ABPs alter the fluctuations of monomer configurations within an actin filament
Hirotaka Ito[1], Kohei Monma[1], Sakura Maesato[1], Kenji Kobayashi[1], Ryoki Ishikawa[2], Hazime Honda[1] ([1]*Dept. of Bioneg., Nagaoka Univ. Tech*, [2]*Gunma Pref. Col. Health Sci.*)

3Pos131 **変異リアノジン受容体における分子動力学解析とカルシウムシグナル可視化解析の相関**
Correlation of molecular dynamics analysis and calcium signaling in mutant ryanodine receptors
Toshiko Yamazawa[1], Takashi Murayama[2], Hideto Oyamada[3], Junji Suzuki[4], Nagomi Kurebayashi[2], Kazunori Kanemaru[4], Maki Yamaguchi[1], Shigeru Takemori[1], Katsuji Oguchi[3], Takashi Sakurai[2], Masamitsu Iino[4] ([1]*Dept Mol. Physiol., Jikei Univ. Sch. Med.*, [2]*Dept. Pharmacol., Juntendo Univ. Sch. Med.*, [3]*Dept. Pharmacol., Sch. Med., Showa Univ.*, [4]*Dept. Pharmacol., Grad. Sch. Med., The Univ. Tokyo*)

3Pos132 **再構成系を用いた WAVE 複合体制御分子機構の解明**
Reconstitution of the WAVE complex regulation mechanism
Tomotaka Komori[1,2], Scott Hansen[2], R. Dyche Mullins[2] ([1]*Univ. of Tokyo*, [2]*UCSF*)

3Pos133 **超解像イメージングを用いた単離ミトコンドリアの膜構造観察**
Super-resolution Imaging of Isolated Mitochondria with Structured Illumination Microscopy
Takahiro Shibata[1], Saki Yamashita[1], Kaoru Katoh[2], Yoshihiro Ohta[1] ([1]*Grad. Sch. Life Sci. & Bio Tech., TUAT*, [2]*AIST*)

3Pos134 **機能的な FRET プローブを用いた一細胞における大腸菌走化性受容体活性の検出**
The detection of chemoreceptor cluster's activity in a single *E. coli* cell by the functional FRET probe
Tomoko Horigome[1], Hajime Fukuoka[2], Hiroto Takahashi[3], Yuichi Inoue[3], Akihiko Ishijima[2,3] ([1]*Grad. Sch. Life Sci., Tohoku Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*IMRAM, Tohoku Univ.*)

3Pos135 **SOS を介した Ras positive feedback 制御から見た SOS Noonan 症候群変異体の分類**
Classification of molecular dynamics in SOS Noonan syndrome mutants from the properties of SOS-mediated RAS positive feedback
Yuki Nakamura[1,2], Kayo Hibino[3], Yasushi Sako[1,2] ([1]*RIKEN*, [2]*Osaka university*, [3]*NIG*)

3Pos136 **単一心筋細胞の細胞外電位計測**
Measurement of extracellular potential in single cardiomyocyte
Jyunpei Shimada[1], Kenji Yasuda[2], **Tomoyuki Kaneko**[1] ([1]*LaRC, Dept. Frontier Biosci., Hosei Univ.*, [2]*IBB, TMDU*)

3Pos137 **原子間力顕微鏡によるホヤ初期発生胚の弾性率の時空間測定**
Spatial-temporal change in elastic modulus of ascidian embryo during an early stage of development by atomic force microscopy
Yuki Fujii[1], Wataru Koizumi[2], Kohji Hotta[2], Kotaro Oka[2], Takaharu Okajima[1] ([1]*Grad. Sch. Inform. Sci and Tech., Univ. Hokkaido*, [2]*Grad. Sch. Biosci. and Bioinfo., Univ. Keio*)

3Pos138 **接着分子 CADM1 による膵島 α 細胞グルカゴン分泌調節機構の解明**
Cell adhesion molecule 1 (CADM1) regulate glucagon secretion in pancreatic α cells
Satoru Yokawa[1,2], Ryousuke Oguri[2], Yoshikazu Inoh[2], Ryo Suzuki[2], Tadahide Furuno[2], Naohide Hirashima[1] ([1]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [2]*Sch. Pharm., Aichi Gakuin Univ.*)

3Pos139 **HubP は FlhG を極に局在させることで海洋性ビブリオ菌のべん毛本数を制御する**
HubP regulates the flagellar number by localizing FlhG at the cell pole in marine *Vibrio*
Norihiro Takekawa, Soojin Kwon, Seiji Kojima, Michio Homma (*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*)

3Pos140 **ビブリオ菌 PomB 変異に対するべん毛モーター機能へのセリンの影響**
Effect of serine on the flagellar motor function of a PomB mutant in *Vbrio alginolitycus*
Tatsuro Nishikino, Yasuhiro Onoue, Norihiro Takekawa, Shiwei Zhu, Seiji Kojima, Michio Homma (*Nagoya University Graduate School of Science*)

3Pos141 **細胞性粘菌における高感度膜電位イメージング**
High-sensitivity fluorescence imaging of membrane potential in *Dictyostelium*
Yusuke V. Morimoto[1], Masahiro Ueda[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Sci., Osaka Univ.*)

生体膜･人工膜：構造･特性 / Biological & Artificial membrane: Structure & Property

3Pos142 **固体 NMR と MD シミュレーションによる抗菌ペプチド ボンビニン H2 および H4 の DMPC 膜結合構造の解析**
Membrane binding structure of Bombinin H2 and H4 peptides in DMPC bilayers as studied by solid-state NMR and MD simulation
Izuru Kawamura[1], Yuki Kitahashi[1], Namsrai Javkhlantugs[1,2], Nyamsambuu Altannavch[2], Kazuyoshi Ueda[1], Akira Naito[1] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Natl Univ. Mongolia*)

3Pos143 **オクタアルギニンの膜透過促進効果を示す曲率誘導性ペプチド**
Curvature Inducing Peptides Accelerating Membrane Translocation of Octaarginine (R8)
Tomo Murayama, Shiroh Futaki (*ICR, Kyoto Univ.*)

3Pos144 **ラマン分光によるスフィンゴミエリンの膜分布に関する研究**
Sphingomyelin distribution in model membranes by Raman Spectroscopy
Koichiro Shirota[1], Kiyoshi Yagi[2], Takehiko Inaba[1], Pai-Chi Li[2], Yuji Sugita[2], Toshihide Kobayashi[1] ([1]*LBL, RIKEN*, [2]*TMSL, RIKEN*)

3Pos145 **合成生物学研究のための一枚膜ベシクル内 DNA コンピュータ基盤遺伝子調節システムの開発**
Development of a DNA computer-based gene-regulatory system confined in a giant unilamellar vesicle for synthetic biology research
Koh-ichiroh Shohda[1], Toru Nishikata[1], Yutetsu Kuruma[2], Akira Suyama[1] ([1]*The University of Tokyo*, [2]*Tokyo Institute of Technology*)

3Pos146 長鎖リン脂質と短鎖リン脂質で構成される脂質多成分系の相挙動と構造変化
The phase behavior and the structural changes of lipid multi-component system consisting of long- and short-chain phospholipids
Ryota Kobayashi, Tetsuhiko Ohba (*Dept. of Phys., Tohoku Univ.*)

3Pos147 膜タンパク質の機能構造解析を指向した安定かつサイズ制御可能なナノディスクの開発
Development of stable and size-controllable nanodiscs for biophysical analysis of membrane proteins
Hiroaki Kondo, Keisuke Ikeda, Minoru Nakano (*Grad. Sch. Med. Pharm. Sci., Univ. Toyama*)

3Pos148 アクチン重合によって引き起こされるリポソームの変形に与えるリン脂質組成の影響
Effect of lipid composition on the actin polymerization-driven shape change of giant liposomes
Shunsuke Tanaka, Masahito Hayashi, Kingo Takiguchi (*Div. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

3Pos149 巨大細胞膜小胞をマニピュレーションするための新規デバイスの開発
Development of new device for manipulation of giant plasma membrane vesicles
Keishi Sato[1], Rebun Sakane[2], Takayuki Nakaya[2], Takashi Okuno[3] ([1]*Grad. Sch. Sci. Eng., Univ. Yamagata*, [2]*Namiki Precision Jewel Co., Ltd*, [3]*Fac. Sci., Univ. Yamagata*)

生体膜･人工膜：動態 / Biological & Artificial membrane: Dynamics

3Pos150 抗菌ペプチド・PGLa が誘起する脂質膜中のポア形成とマガイニン２との相乗効果
Antimicrobial Peptide PGLa-Induced Pore Formation in Lipid Membranes and its Synergistic Effect with Magainin 2
Farliza Parvez[1], Jahangir Md. Alam[2], Hideo Dohra[3], Masahito Yamazaki[1,2,4] ([1]*Int. Biosci., Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Electronics, Shizuoka Univ.*, [3]*Res. Inst. Green Sci. Tech., Shizuoka University*, [4]*Dept. Phys., Grad. Sch. Sci., Shizuoka Univ.*)

3Pos151 **Activation energy of the tension-induced pore formation in lipid membranes**
Mohammad Abu Sayem Karal[1], Masahito Yamazaki[1,2] ([1]*Int. Biosci., Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Electronics, Shizuoka Univ.*)

3Pos152 **Effects of Line Tension on Antimicrobial Peptide Magainin 2-Induced Pore Formation**
Jahangir Md. Alam[1], Mohammad Abu Sayem Karal[2], Victor Levadny[3], Masahito Yamazaki[1,2,4] ([1]*Res. Inst. Electronics, Shizuoka Univ.*, [2]*Int. Biosci., Grad. Sch. Sci. Tech., Shizuoka Univ.*, [3]*Theo. Pro. Center Phys.-Chem. Pharm., Rus. Acad. Sci.*, [4]*Dept. Phys., Grad. Sch. Sci., Shizuoka Univ.*)

3Pos153 膜タンパク質-脂質相互作用の定量的解析法の開発と応用
Quantitative analysis of protein-lipid interactions
Takaharu Mori, Yuji Sugita (*RIKEN*)

3Pos154 脂質二重膜の流動性へのグラフェン酸化物の影響
Effect of graphene oxide substrate on the fluidity of lipid bilayer membrane
Yoshiaki Okamoto[1], Toshinori Motegi[2], Seiji Iwasa[1], Adarsh Sandhu[2], Ryugo Tero[1,2,3] ([1]*Dept. Environmental and Life Sci., Toyohashi Univ. Tech.*, [2]*EIIRIS, Toyohashi Univ. Tech.*, [3]*CREST, JST*)

生体膜･人工膜：興奮･チャネル / Biological & Artificial membrane: Excitation & Channels

3Pos155 **Conformational Control of Voltage Sensor Domains**
Morten Bertz, Kazuhiko Kinosita, Jr. (*Waseda University, Dpt. of Physics*)

3Pos156 電位依存性プロトンチャネルの 2 量体と単量体間のゲーティング特性の違いを増大させる変異体の解析
Mutation of a hydrophobic residue in S4 enhances the difference between monomeric and dimeric voltage-gated proton channels
Akira Kawanabe, Yasushi Okamura (*Osaka University*)

3Pos157 イオン透過と選択性を記述する速度論モデル
A kinetic model describing punch-through of Na^+ through KcsA potassium channel
Takashi Sumikama, Kenichiro Mita, Shigetoshi Oiki (*Univ. of Fukui*)

生体膜･人工膜：輸送 / Biological & Artificial membrane: Transport

3Pos158 **Proton transfer between cytochrome oxidases and the ATP synthase: examining the role of the membrane environment**
Duncan McMillan[1,2], Sophie Marritt[3], Mengqui Li[2], Sune Jorgensen[4], Rikya Watanabe[1], Nikos Hatzakis[4], Julea Butt[3], Lars Jeuken[2], Gregory Cook[5], Hiroyuki Noji[1] ([1]*Department of Applied Chemistry, University of Tokyo, Tokyo, Japan*, [2]*School of Biomedical Sciences, University of Leeds, Leeds, UK*, [3]*School of Chemistry, Norwich Research Park, University of East Anglia, UK*, [4]*Nanoscience Center, University of Copenhagen, Copenhagen, Denmark*, [5]*Department of Microbiology and Immunology, University of Otago, Dunedin, New Zealand*)

3Pos159 反転膜ベシクルを用いたべん毛軸構造の構築
Construction of the flagellar axial structure using inverted membrane vesicles
Chinatsu Tatsumi[1], Hiroyuki Terashima[1], Tohru Minamino[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci., Univ. Osaka*, [2]*Grad. Sch. Frontier Biosciences., Univ. Osaka*)

3Pos160 リン脂質輸送タンパク質 Sec14 のホスファチジルコリン輸送に対する脂質膜組成の影響
Effect of membrane-components on Sec14-mediated phosphatidylcholine transfer
Taichi Sugiura, Keisuke Ikeda, Minoru Nakano (*Grad. Sch. Med. Pharm. Sci., Univ. Toyama*)

神経科学·感覚 / Neuroscience & Sensory systems

3Pos161 **線虫（C. elegans）がかぎ分けられるにおい濃度差を行動と神経活動から明らかにする**
Specific behavior and neural activity of Caenorhabditis elegans unveil how exactly it detects difference of odor concentration
Hisashi Shidara, Masanari Makino, Kohji Hotta, Kotaro Oka (*Grad. Sch. Sci. and Tech., Keio Univ.*)

3Pos162 **視覚ー味覚条件付けによるモノアラガイの視覚特性の検討**
Experimental study about the visual characteristics of a pond snail, *Lymnaea stagnalis*, by visual-appetitive conditioning
Satoshi Takigami[1], Momoko Koide[2], Tetsuro Horikoshi[2] ([1]*Grad. Sch. Bio., Tokai Univ.*, [2]*Dept. Biomed. Eng., Sch. Engineering, Tokai Univ.*)

3Pos163 **カエル神経筋接合部シナプスでの短期可塑性の二項分布解析: 促通では放出可能な小胞数が増加し、増強では放出確率が増加する**
Binomial distribution analysis of short-term plasticity, facilitation and potentiation, at the frog NMJ: n and p increases, respectively
Naoya Suzuki (*Dept. Phys., Sch. Sci., Nagoya Univ.*)

神経回路·情報処理 / Neuronal circuit & Information processing

3Pos164 **培養神経回路網における神経活動パターンの時間遷移の解析**
Analysis of the transition of electrical activity patterns in cultured neuronal cells
Takumi Okada, Keisuke Izutani, Hidekatsu Ito, Wataru Minoshima, Suguru N. Kudoh (*Sch. Sci. & Tech., Kwansei Gakuin Univ.*)

3Pos165 **やわらかいボディのダイナミクスを用いて情報処理を実装する**
Information Processing Using Soft Body Dynamics
Kohei Nakajima[1,2] ([1]*The Hakubi Center for Advanced Research, Kyoto University*, [2]*Graduate School of Informatics, Kyoto University*)

3Pos166 **ランビエ絞輪近傍の BK チャネルは軸索の高頻度発火を制御する**
Paranodal BK channels regulate high frequency firing in myelinated axons
Moritoshi Hirono, Hiroaki Misonou (*Grad Sch Brain Sci, Doshisha Univ*)

光生物：視覚·光受容 / Photobiology: Vision & Photoreception

3Pos167 **In-situ 光照射固体 NMR による ppR/pHtrII 複合体の光反応過程の解析**
Hotoreaction pathway of ppR/pHtrII as revealed by in-situ photo irradiation solid-state NMR
Yoshiteru Makino[1], Yuya Tomonaga[1], Tetsurou Hidaka[1], Izuru Kawamura[1], Takashi Okitsu[2], Akimori Wada[2], Yuki Sudo[3], Naoki Kamo[3], Akira Naito[1] ([1]*Grad. Sch. Eng, Yokohama Natl Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Grad. Sch. Med, Dent, Pharm, Okayama Univ.*, [4]*Grad. Sch. Life Sci , Hokkaido Univ.*)

3Pos168 **小角 X 線溶液散乱によるアレスチンとロドプシンの相互作用の解析**
Interaction between visual arrestin and membrane-embedded rhodopsin studied by solution small-angle X-ray scattering
Yasushi Imamoto[1], Kojima Keiichi[1], Toshihiko Oka[2,3], Ryo Maeda[1], Yoshinori Shichida[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Fac. Sci., Shizuoka Univ.*, [3]*Research Inst. Electronics, Shizuoka Univ.*)

3Pos169 **シロイヌナズナ phototropin 2 のＸ線小角散乱による構造研究**
Small-angle X-ray scattering study of Arabidopsis phototropin 2
Mao Oide[1,2], Koji Okajima[1,2], Yuki Sekiguchi[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Takaaki Hikima[2], Masaki Yamamoto[2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*)

3Pos170 **ロドプシンはラフト親和性の短寿命ナノドメインを形成しながら拡散している**
Rhodopsin diffuses in disc membranes by forming raftophilic and transient nanodomains
Fumio Hayashi[1], N. Saito[1], Y. Tanimoto[2], K. Morigaki[2], K. Seno[3] ([1]*Grad. Sch. Sci. Univ. Kobe*, [2]*Grad. Sch. Agri. Univ. Kobe*, [3]*Univ. Sch. Med. Hamamatsu*)

3Pos171 **トランスデューシン活性化能を有するロドプシン褪色中間体と Meta II 中間体の同一性に関する検討**
Does Meta II correspond to R*, the bleaching intermediate activating transducin?
Shuji Tachibanaki[1,2], Ryota Kumakura[1], Whei-Ee Tang[1], Yoichiro Fukunaga[1], Satoru Kawamura[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*)

3Pos172 **Ab initio MD シミュレーションによる PYP の活性部位水素結合ネットワークの解析**
Ab initio MD study on the dynamic structure of the hydrogen bond network in the active site of PYP
Yusuke Kaneta, Hiroshi Watanabe, Tadaomi Furuta, **Minoru Sakurai** (*Center for Biol. Res. & Inform., Tokyo Tech*)

3Pos173 **PYP - Phytochrome Related Protein の発色団が構造へ与える影響**
Effects of chromophores of PYP - Phytochrome Related Protein on the structure
Keito Yoshida, Kento Yonezawa, Yoichi Yamazaki, Mikio Kataoka, Hironari Kamikubo (*Grad. Sch. Mat. Sci., NAIST*)

3Pos174 **液体中ナノ空間拘束下の量子効率増強：色素分子の光学的ホールバーニング過程**
Enhancement of quantum efficiency in a nanometer-sized confinement in liquids: optical hole-burning processes of dye molecules
Hiroshi Murakami (*KPSI, JAEA*)

3Pos175 **Isotope-labeled DNA substrate revealed site-specific interaction with CPD photolyase**
I M. Mahaputra Wijaya[1], Tatsuya Iwata[1], Toshihiko Hamamura[2], Junpei Yamamoto[2], Kenichi Hitomi[3], Shigenori Iwai[2], Elizabeth D. Getsoff[3], Hideki Kandori[1] ([1]*Dept. of Frontier Materials, Nagoya Institute of Technology, Japan*, [2]*Grad. Sch. of Engineering Science, Osaka University, Japan*, [3]*Dept. of Integrative Structural and Computational Biology and The Skaggs Institute for Chemical Biology, The Scripps Research Institute, USA*)

3Pos176 **Crystallographic study of the LM intermediate of squid rhodopsin**
Midori Murakami, Tsutomu Kouyama (*Grad. Sch. Sci., Nagoya Univ.*)

光生物：光合成 / Photobiology: Photosynthesis

3Pos177 **光合成のマルチ時間スケールダイナミクスに対するシステム解析**
Systems approach to the multi-timescale dynamics of photosynthesis
Takeshi Matsuoka[1], Shigenori Tanaka[2], Kuniyoshi Ebina[3] ([1]*KOZO KEIKAKU ENGINEERING Inc.*, [2]*Grad. Sch. Sys. Info., Kobe Univ.*, [3]*Grad. Sch. Hum. Dev. and Env., Kobe Univ.*)

3Pos178 **ホウレン草の PSII 反応中心に生成する光電荷分離状態の時間分解 EPR**
Time resolved EPR study on photoinduced primary charge-separated state of the PSII reaction center from spinach
Masashi Hasegawa[1], Takahiro Sakai[2], Hiroki Nagashima[2], Takashi Tachikawa[1], Hiroyuki Mino[2], Kobori Yasuhiro[1] ([1]*Grad. Sch. Sci., Kobe Univ.*, [2]*Grad. Sch. Sci., Nagoya Univ.*)

3Pos179 **Thermochromatium tepidum 由来 LH1-RC 複合体における耐熱化の分子機構：同位体置換体の FTIR 分析**
Molecular mechanisms for the enhanced thermal stability of LH1-RC complex from Thermochromatium tepidum: isotope-edited FTIR spectroscopy
Yukihiro Kimura[1], Yuki Yura[1], Li Yong[1], Seiu Otomo[2], Takashi Ohno[1] ([1]*Grad. Sch. Agri. Sci., Kobe Univ.*, [2]*Fac. Sci., Ibaraki Univ.*)

3Pos180 **Formation of complexes between genetically modified photosystem I and single-walled carbon nanotubes**
Daisuke Nii[1], Yuichiro Shimada[2], Akane Hosokawa[2], Yosuke Nozawa[3], Masahiro Ito[1], Yoshikazu Homma[1], Tatsuya Tomo[1] ([1]*Department of Physics, Graduate School of Science, Tokyo University of Science*, [2]*Department of Industrial Chemisty, Tokyo University of Science*, [3]*Department of Physics, Tokyo University of Science*)

3Pos181 **光合成水分解 Mn_4CaO_5 クラスターの光活性化における Mn^{2+}結合部位の ATR-FTIR 検出**
ATR-FTIR detection of the Mn^{2+} binding site in photoactivation of the water-oxidizing Mn_4CaO_5 cluster in photosystem II
Akihiko Sato, Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Univ. Nagoya*)

3Pos182 **光化学系 II の部位特異的変異導入によるクロロフィル二量体 P680 の電子構造の**
Electronic structure of the chlorophyll dimer P680 modified by site-directed mutation at a nearby amino acid residue in photosystem II
Motoki Yamaguchi, Ryo Nagao, Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

放射線生物学･活性酸素 / Radiobiology & Active oxygen

3Pos183 **重イオン線照射によってヒト正常線維芽細胞に誘導されるバイスタンダーシグナル**
Heavy-ion induced bystander signaling in normal human fibroblasts
Masanori Tomita[1], Hideki Matsumoto[2], Tomoo Funayama[3], Yuichiro Yokota[3], Kensuke Otsuka[1], Munetoshi Maeda[1,4], Yasuhiko Kobayashi[3] ([1]*Radiat. Safety Res. Cent., CRIEPI*, [2]*Biomed. Imaging Res. Cent., Univ. Fukui*, [3]*Radiat. Biol. Res. Div., QuBS, JAEA*, [4]*R&D, WERC*)

3Pos184 **超音波によるケージド化合物の活性化のメカニズム**
Mechanism of activation of caged compounds induced by ultrasound
Haruko Koura[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Mater. & Human Env. Sci., Shonan Inst. of Tech.*)

生命の起源･進化 / Origin of life & Evolution

3Pos185 **凍結融解を用いたリポソーム融合手法の確立と継続的な RNA 複製反応を伴う人工細胞増殖システムの構築**
Reconstitution of artificial cell growth by coupling the RNA replication and propagation in liposomes using a freeze-thawing method
Gakushi Tsuji[1], Satoshi Fujii[2], Takeshi Sunami[3], Tetsuya Yomo[1,2] ([1]*Fbs, Osaka University*, [2]*IST, Osaka University*, [3]*IAI, Osaka University*)

3Pos186 **大腸菌クローン集団の抗生物質に対する不均一な応答**
Heterogeneous Responses to Antibiotic stress in a Clonal Population of *Escherichia coli*
Miki Umetani[1], Yuichi Wakamoto[2,3], Chikara Furusawa[1] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Arts and Sci., Univ. of Tokyo*, [3]*Research Center for Complex Systems Biology*)

ゲノム生物：ゲノム構造 / Genome biology: Genome structure

3Pos187 **細胞外刺激前後のクロマチンダイナミクスの変化の 1 分子イメージング解析**
Single molecule imaging analysis of chromatin dynamics in response to extracellular stimulation
Kayo Hibino[1,2], Ryosuke Nagashima[1,2], Kazuhiro Maeshima[1,2] ([1]*NIG*, [2]*SOKENDAI*)

バイオインフォマティクス：ゲノム構造 / Bioinformatics: Structural genomics

3Pos188 **タンパク質における分子トンネルの形状分析**
A Method for Detecting and Analyzing Shapes of Molecular Tunnels in Proteins
Midori Yano[1], Kei Yura[1,2] ([1]*Grad. Sch. Hum. Sci., Univ. Ochanomizu*, [2]*NIG*)

3Pos189 **剛体アンサンブルドッキングによって得られた候補構造群における相互作用残基ペアの特徴の解析**
Analysis of background interaction properties of protein complexes generated by rigid-body ensemble docking
Nobuyuki Uchikoga[1], Yuri Matsuzaki[2], Masahito Ohue[3], Yutaka Akiyama[2,3] ([1]*Dept. of Physics, Chuo Univ.*, [2]*ACLS, Tokyo Tech*, [3]*Grad. Sch. of Inform. Sci. and Eng, Tokyo Tech*)

3Pos190 **アミノ酸プロファイルによるタンパク質ペプチド複合体のポストドッキング解析**
Post-docking analysis of protein-peptide complex structure using amino acid profiles
Masahito Ohue[1], Nobuyuki Uchikoga[2], Yuri Matsuzaki[3], Yutaka Akiyama[1,3] ([1]*Dept. Comput. Sci., Grad. Sch. Inform. Sci. Eng., Tokyo Tech.*, [2]*Dept. Phys., Facul. Sci. Eng., Chuo Univ.*, [3]*ACLS, Tokyo Tech.*)

3Pos191 **タンパク質構造の持つトポロジーの安定性を評価する**
How to estimate topological stability of protein structures
Shintaro Minami[1], George Chikenji[2], Motonori Ota[1] ([1]*Dept. of Comp. Sci., Nagoya Univ.*, [2]*Dept. of Eng., Nagoya Univ.*)

3Pos192 **β-Trefoil タンパクのフォールディングコアの残基間平均距離統計に基づく予測**
Prediction of folding nuclei of beta-Trefoil proteins based on the inter-residue average distance statistics
Takuya Kirioka, Norihiro Kanemaru, Takeshi Kikuchi (*Ritsumeikan University*)

バイオインフォマティクス：ゲノム機能 / Bioinformatics: Functional genomics

3Pos193 **サポートベクターマシンを用いたアミノ酸配列からの脂質結合予測および脂質結合残基予測**
Predicting lipid-binding proteins and lipid-binding residues from amino acid sequences by using support vector machine
Kokoro Ueki, Shugo Nakamura, Kentaro Shimizu (*Grad Sch. of Agr., The Univ. of Tokyo*)

生態･環境 / Ecology & Environment

3Pos194 **少数個体がつくる鮎の群れに内在する相互作用構造**
Interaction Structure of fish schools in a small population
Takayuki Niizato[1], Hisashi Murakami[2], Kazuki Sangu[1], Yuta Nishiyama[3], Kohei Sonoda[4], Yukio Gunji[2] ([1]*Tsukuba University*, [2]*Waseda University*, [3]*Osaka University*, [4]*Shiga University*)

数理生物 / Mathematical biology

3Pos195 **マイクロアレイデータに基づく植物の遺伝子発現揺らぎと機能の関係**
Analysis of relationship between noise of gene expression and function of plants based on microarray data
Kodai Hirao[1], Atsushi Nagano[2], Akinori Awazu[1,3] ([1]*Dept. of Mathematical and Life Sciences, Hiroshima Univ*, [2]*Dept. of Plant Life Sciences, Ryukoku Univ*, [3]*RcMcD, Hiroshima Univ*)

3Pos196 **3 次元細胞シミュレーションにおける高次精度・高解像度スキームの評価**
Evaluation of higher order and high resolution schemes for 3D cell simulation
Chikara Sawa[1], Masakazu Tanaka[1], Hayato Takeuchi[1], Kiminori Toyooka[2], **Eisuke Chikayama**[1,3,4] ([1]*Niigata Univ. Inter. Infor. Stud.*, [2]*Mass Spec. Micros. Unit, RIKEN*, [3]*Env. Metab. Aanal. Res. Team, RIKEN*, [4]*Image Proc. Res. Team, RIKEN*)

3Pos197 **分子の種類を識別する細胞情報処理の確率モデルとその情報論的考察**
Stochastic modeling and information-theoretical study of molecular discrimination
Masashi K. Kajita[1], Kazuyuki Aihara[1,2], Tetsuya J. Kobayashi[2] ([1]*Grad. Sch. Inf. Sci. Tech., Univ. Tokyo*, [2]*IIS, Univ. Tokyo*)

3Pos198 **On thermodynamics of macromolecular association in vivo: Theoretical and simulation studies with a coarse-grained model**
Tadashi Ando[1], Yuji Sugita[1,2,3,4], Michael Feig[5] ([1]*RIKEN QBiC*, [2]*RIKEN AICS*, [3]*RIKEN iTHES*, [4]*RIKEN TMS*, [5]*Michigan State Univ.*)

3Pos199 **Clausius Inequality in Population Growth**
Yuki Sugiyama, Tetsuya J. Kobayashi (*IIS, Univ. Tokyo*)

非平衡･生体リズム / Nonequilibrium state & Biological rhythm

3Pos200 **バクテリアケモスタットのためのマイクロ流体デバイスの構築**
Development of microfluidic device for bacterial chemostat
Manami Ito[1], Haruka Sugiura[1], Shotaro Ayukawa[1,2], Daisuke Kiga[1,3], Masahiro Takinoue[1,4] ([1]*Dept. Comput. Intell. Syst. Sci., Tokyo Tech.*, [2]*ACLS, Tokyo Tech.*, [3]*ELSI, Tokyo Tech.*, [4]*PRESTO, JST*)

3Pos201 **匂いを嗅ぎ走り出す走化性液滴：ガス刺激からの逃避行動への応答**
Chemotactic behavior of a liquid droplet: Smelling and escaping against evil gas
Hiroki Sakuta[1], Nobuyuki Magome[2], Yoshihito Mori[3], Kenichi Yoshikawa[1] ([1]*Facul. Lif. Med. Sci., Doshisha Univ.*, [2]*Chem., Dokkyo Med. Univ.*, [3]*Facul. Sci., Ochanomizu Univ.*)

計測 / Measurements

3Pos202 **In situ マイクロ波照射 NMR 法を用いた生体系におけるマイクロ波効果の解析**
Analysis of microwave effects on biological system by in situ microwave irradiation NMR spectroscopy
Yugo Tasei[1], Takuya Yamakami[2], Fumikazu Tanigawa[1], Izuru Kawamura[1], Teruaki Fujito[3], Kiminori Ushida[2], Motoyasu Sato[4], Akira Naito[1] ([1]*Graduate School of Engineering, Yokohama National University*, [2]*Department of Chemistry, School of Science, Kitasato University*, [3]*Probe Laboratory Inc.*, [4]*Faculty of Engineering, Chubu University*)

3Pos203 **光制御水素化アモルファスシリコン薄膜上の化学反応性積層ゲルを用いた植物由来分子の検出**
Detection of plant derived molecules using electrochemical laminated gels photo-controlled on hydrogenated amorphous silicon film
Shotaro Minato[1], Yutaka Tsujiuchi[1], Hiroshi Masumoto[2], Takashi Goto[3] ([1]*Material Science and Engineering, Akita University*, [2]*Frontier Research Institute for Interdisciplinary Sciences, Tohoku University*, [3]*Institute for Materials Research, Tohoku University*)

3Pos204 **バクテリアプロトプラストとマイクロ膜チャンバーの融合細胞内部の ATP 濃度の測定**
Measurement of ATP concentration in fusion cell of bacterial protoplast into micro-sized lipid membrane chamber
Hiroki Ashikawa[1], Kazuhito Tabata V.[1,2], Rikiya Watanabe[1,3], Hideyuki Yaginuma[3,4], Hiroyuki Noji[1,3] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*PRESTO, JST*, [3]*CREST, JST*, [4]*QBiC, RIKEN*)

3Pos205 **超高感度 ELISA の開発：血中 HIV-1 p24 ならびに尿中アディポネクチンの測定**
Ultrasensitive ELISA: Detection of HIV-1 p24 in blood and that of adiponectin in urine at attomole level
Etsuro Ito (*Kagawa Sch. Pharmaceu. Sci., Tokushim Bunri Univ.*)

3Pos206 **原子間力顕微鏡による力学的単一細胞診断：細胞力学量のばらつきの空間依存性**
Single cell mechanical diagnostics using atomic force microscopy: how cell-to-cell mechanical variation depends on the cell position
Maki Sawano[1], Kaori Shigetomi(Kuribayashi)[1], Kinho Shu[1], Ryosuke Takahashi[1], Agusu Subagyo[2], Kazuhisa Sueoka[2], Ryosuke Tanaka[1], Takaharu Okajima[1] ([1]*Grad. Sch. bio., Univ. Hokkaido*, [2]*Grad. Sch. Nano., Univ. Hokkaido*)

バイオイメージング / Bioimaging

3Pos207 **Development of target-specific single-dot chemical probes in live cells via intracellular click reaction**
Yanyan Hou[1], Satoshi Arai[1], Tetsuya Kitaguchi[1,2], Madoka Suzuki[1,2] ([1]*WASEDA Biosci Res Inst Singapore (WABIOS)*, [2]*Org Univ Res Initiatives, Waseda Univ*)

3Pos208 **一分子観察のための高速 AFM /チップ増強蛍光顕微鏡複合機**
Combined system of HS-AFM and tip-enhanced fluorescence microscopy for single molecular imaging
Shingo Fukuda[1], Takayuki Uchihashi[1,2], Ryota Iino[3], Toshio ANdo[1,2] ([1]*Sch. Math. & Phys., Col. Sci. & Eng., Kanazawa Univ.*, [2]*Bio-AFM FRC, Inst. Sci. & Eng., Kanazawa Univ*, [3]*Okazaki Inst. Integr. Biosci., NINS*)

3Pos209 **仮想電子顕微鏡 VEM の開発**
Development of virtual electron microscopy to supply unified user interface
Ayaka Iwasaki (*Kyushu Institute of Technology*)

3Pos210 **大量データ計測技術を用いたシアノバクテリアの低温コヒーレントＸ線回折イメージング**
Cryogenic coherent X-ray diffraction imaging of cyanobacteria using the high-throughput measurement techniques
Amane Kobayashi[1,2], Yuki Sekiguchi[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Yayoi Inui[3], Sachihiro Matsunaga[3], Yuki Takayama[2], Masaki Yamamoto[2] ([1]*Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*, [3]*Sci. Tech., Tokyo Univ. Sci.*)

3Pos211 **細胞内リン酸化タンパク質の多並列超迅速解析法**
Multi-parallel super rapid analysis methods of intracellular phosphoproteins
Naoki Takeuchi[1], Miho Suzuki[1], Akira Kurisaki[2], Koichi Nishigaki[1] ([1]*Grad. Sch. of Sci. and Eng., Saitama Univ.*, [2]*AIST*)

3Pos212 **シグナル分子の時空間的に異なる局在が走化性を調節している**
Spatiotemporal different localizations of multiple signaling molecules mediate chemotaxis in Dictyostelium cells
Yuki Tanabe[1,2], Masahiro Ueda[1,2] ([1]*Graduate school of Science, Osaka University*, [2]*QBiC, RIKEN*)

3Pos213 **CLIP-170 phosphorylation mediates repositioning of microtubule-organizing center during T cell activation**
Wei Ming Lim, Yuma Ito, Makio Tokunaga, Kumiko Sakata-Sogawa (*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*)

3Pos214 **リポ多糖刺激における炎症抑制タンパク質 PDLIM2 活性化のイメージング定量解析**
Quantitative imaging analysis of anti-inflammatory protein PDLIM2 activation upon LPS stimulation
Shota Ichikawa[1], Yuma Ito[1], Takashi Tanaka[2], Makio Tokunaga[1], Kumiko Sakata-Sogawa[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*IMS-RCAI, RIKEN*)

3Pos215 **温度上昇に伴うマクロファージ運動の活性化**
Activation of motility of macrophage at temperature jump
Hideo Saitou, Masamine Shintani, Sayaka Kita, Hideo Higuchi (*The university of tokyo*)

3Pos216 **高感度および高解像度電気化学チップデバイスによる幹細胞の機能イメージング**
Electrochemical imaging of stem cell function using high-sensitivity and high-resolution electrochemical chip devices
Yusuke Kanno[1], Kosuke Ino[1], Hitoshi Shiku[1], Tomokazu Matsue[1,2] ([1]*Graduate School of Environmental Studies, Tohoku Univ.*, [2]*WPI-Advanced Institute for Materials Research, Tohoku Univ.*)

3Pos217 **ゼブラフィッシュグリア細胞活動の In vivo イメージング**
In vivo imaging of glial cell activity in zebrafish
Hiroko Bannai, Masashi Tanimoto, Shigeo Sakuragi, Yurie Matsutani, Yoichi Oda (*Grad. Sch. Biol. Sci., Univ. Nagoya*)

3Pos218 **VSFG 検出赤外超解像顕微分光法による羽毛 β-ケラチンの分子配向赤外イメージング**
Orientation-sensitive IR imaging of feather β-keratins by a VSFG-detected IR super-resolution micro-spectroscopy
Yukihisa Watase, Kohei Ushio, Masaaki Fujii, Makoto Sakai (*Chem. Res. Lab, Tokyo. Tech*)

バイオエンジニアリング / Bioengineering

3Pos219 **マイクロ波照射下での酵素反応の出力依存性に関する研究**
The study of microwave output dependence on enzymatic reaction under microwave irradiation
Fujiko Aoki[1], Kenshi Haraguchi[2], Arata Shiraishi[1], Syokichi Ohuchi[1] ([1]*Dept. Lifesci. & Syst. Eng., Kyushu Inst. Tech.*, [2]*Dept. Biosci. & Bioinform., Kyushu Inst. Tech.*)

3Pos220 **弾性率可変マイクロファイバーゲルマトリックスにおけるがん細胞の三次元運動表現型評価**
Phenotypic differences in 3D movement of tumor cells observed in the microfiber gel matrices with tunable elasticity
Yu Nakamura, Satoru Kidoaki (*Institute for Materials Chemistry and Engineering, Kyushu Univ.*)

3Pos221 **cDNA display 法によるリポソーム結合ペプチドの試験管内進化**
In Vitro Selection of Liposome Anchoring Peptide by cDNA display
Naoto Nemoto, Ryouya Okawa, Yuki Yoshikawa, Toshiki Miyajima, Shota Kobayasi (*Grad. Sch. of Sci. and Eng., Saitama University*)

3Pos222 **Clogging of DNA driven through a nano-scale pore or slit**
Naoto Sakashita, Yuta Kato, Yoshitaka Tanida, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

3Pos223 **明視野/蛍光マルチイメージングフローサイトメーターを用いた形状情報認識を用いた細胞識別技術の検討**
Studies on identification of cells using visible morphological information using bright field/fluorescent multi-imaging flow cytometer
Akihiro Hattori[1], Hyonchol Kim[1], Hideyuki Terazono[2], Masao Odaka[1], Kenji Matsuura[1], Mathias Girault[1], Kenji Yasuda[1,2] ([1]*Kanagawa Academy of Science and Technology*, [2]*Tokyo Medical and Dental University*)

3Pos224 **Multiple-viewpoint analysis of diversity in T cell receptors**
Ryo Yokota[1,2], Yuki Kaminaga[3], Tetsuya J. Kobayashi[1,2,3] ([1]*Inst. Ind. Sci., Univ. Tokyo*, [2]*Res. & Edu. Platf. Dyn. Liv. States*, [3]*Sch. Eng., Univ. Tokyo*)

3Pos225 **DNA とナノポアを用いたマイクロメディカルドロップレットシステムの開発**
Micro-Medical-Droplet system using DNA and biological nanopore
Moe Hiratani, Masayuki Ohara, Ryuji Kawano (*Tokyo Univ. of Agr. and Tech.*)

3Pos226 **モータータンパク質で駆動する運動界面の構築と細胞の力学刺激**
Dynamic substrate driven by motor proteins for mechanical cellular stimuli
Ryuzo Kawamura, Daiki Uehara, Naritaka Kobayashi, Seiichiro Nakabayashi, Hiroshi Yoshikawa (*Dept. Chem., Saitama Univ.*)

3Pos227 **ポリビニルアルコールゲルで固体化されたバクテリオロドプシンの機能・構造特性に関する分光学的研究**
Structural and Functional Characteristics of Bacteriorhodopsin Immobilized with Poly(Vinyl Alcohol) Gel Studied by Spectroscopic Methods
Hikaru Tanaka[1], Yasunori Yokoyama[1], Masashi Sonoyama[2], Koshi Takenaka[1] ([1]*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*, [2]*Div. Mol. Sci., Fac. Sci. Tech., Gunma Univ.*)

その他 / Miscellaneous topics

3Pos228 **Development of multiple time step integrators in isothermal and isobaric conditions for efficient MD simulations of biological systems**
Jaewoon Jung[1,3], Tadashi Ando[2], Yasuhiro Matsunaga[1], Yuji Sugita[1,2,3,4] ([1]*RIKEN AICS*, [2]*RIKEN QBiC*, [3]*RIKEN TMS*, [4]*RIKEN iTHES*)

3Pos229 **フォトクロミック分子を使った紫外光による細胞死の反応機構**
Reaction mechanism on cell death due to photochromic molecules upon UV irradiation
Satoshi Yokojima[1,4], Ryuhei Kodama[2], Kimio Sumaru[3], Shinichiro Nakamura[4], Kingo Uchida[2] ([1]*Tokyo Univ. Pharmacy and Life Sci.*, [2]*Ryukoku Univ.*, [3]*AIST*, [4]*Riken*)

3Pos230 **Enhanced efflux activity assists E. coli antibiotic tolerance**
Yingying Pu[1], Zhilun Zhao[1], Yingxing Li[1], Jin Zhou[1], Qi Ma[1], Yuehua Ke[1], Yun Zhu[1], Huiyi Chen[2], Hao Ge[1], Yujie Sun[1], Xiaoliang Sunney Xie[1,2], Fan Bai[1] ([1]*Biodynamic Optical Imaging Center, Peking University, Beijing, China*, [2]*Department of Chemistry and Chemical Biology, Harvard University, Cambridge, MA, USA*)

1SAA-01 細菌べん毛モーターの力学特性
Dynamics of the nano-rotary motor of bacterial flagella

Yoshiyuki Sowa (*Dept. of Frontier Bioscience, Hosei Univ.*)

Many species of bacteria swim towards their favorable conditions, propelled by rotating their flagella, which extend from the cell body. Each helical flagellum turned like a screw by a tiny (~50 nm in diameter) rotary molecular motor at its base. The motor consist of a rotor ring and ~10 independent stator units. It spins at several hundreds Hz in both direction, driven by ion flux across the cytoplasmic membrane. By applying high-resolution optical microscopy to the motor, the elementary processes of its rotation were detected. We recently showed the dynamics and assembly of the motor controlled automatically in response to external conditions. In this talk, I will present the experimental studies of the dynamics of nano-rotary motor of bacterial flagella.

1SAA-02 先進光学顕微鏡により明らかになる繊毛とアーキアべん毛の力学機能
Mechanical function of cilia and archaeal flagella resolved by advanced optical microscopes

Takayuki Nishizaka (*Gakushuin Univ.*)

In our research group, various types of advanced optical microscopy have been developed to focus, mainly at the single molecular level, on studies of how biomolecules work. These techniques including 3-D tracking and optical trap were applied to subjects of super-molecular assembly: beating of mouse tracheal cilia; and swimming motility of archaea. Mechanical properties of the single units, a permeabilized cilium immobilized on the glass and a sole bundle of flagella of swimming halophilic archaeon, were characterized with multiple parameters, such as frequency of beating and rotation, moving handedness through 3-D plots, numerical estimation of drag and torque. The accurate quantification of the parameters enables to establish possible models of molecular machineries.

1SAA-03 Regulation of mammalian flagellar and ciliary motility by post-translational modifications of axonemal microtubule

Koji Ikegami, Mitsutoshi Setou (*Hamamatsu Univ. Sch. Med.*)

The axoneme of flagella and cilia is composed of microtubule, which undergoes post-translational modifications, polyglutamylation and polyglycylation. These modifications are highly unique forms of modification where tens of glutamates or glycines are attached as a branched polypeptide on the surface of microtubule. The long branched poly-glutamate or -glycine chain is thought to affect ciliary and flagellar motility. In the symposium, we discuss different effects of polyglutamylation and polyglycylation on, especially mammalian, ciliary and flagellar motility, presenting macroscopic and microscopic phenotypes of ciliary and flagellar beating pattern affected in mice deficient in modifications-performing enzymes.

1SAA-04 マウス胚ノード繊毛においては中心構造体の欠損が回転運動パターンを可能とさせるがその代償として構造配置の不安定性をまねく
The absence of radial spokes allows rotational movement but confers ultrastructural instability in mouse node cilia

Kyosuke Shinohara[1], Duanduan Chen[2], Tomoki Nishida[1], Kazuyo Misaki[3], Shigenobu Yonemura[3], Hiroshi Hamada[1] ([1]*Osaka University*, [2]*Bejing Institute of Technology*, [3]*RIKEN*)

Determination of left-right asymmetry in mouse embryos is established by a leftward flow that is generated by clockwise rotation of node cilia. Here we show exposure to the microtubule-stabilizing drug paclitaxel markedly changes the motion pattern and ultrastructure of node cilia. In vivo observations and a computer simulation revealed that a regular 9+0 arrangement of doublet microtubules is essential for unidirectional rotation of node cilia. The 9+2 motile cilia of the airway, which manifest planar beating, are resistant to Taxol treatment. However, airway cilia of mice lacking the radial spoke head undergo rotational movement are sensitive to Taxol. Our results suggest that the absence of radial spokes allows node cilia to rotate but renders them ultrastructurally.

1SAA-05 鞭毛繊毛ダイニンの多様性と運動の可変性
Diversity of axonemal dyneins and variability of their motor properties in different experimental systems

Ritsu Kamiya (*Department of Life Science, Gakushuin Univ.*)

Chlamydomonas flagella have 3 distinct heavy chains (HCs) in the outer dynein arm and 8 major HCs in the inner dynein arm. Previous studies indicated that different HCs produce microtubule sliding at different speeds. However, the velocities measured in different experimental systems varied greatly, leading us to speculate that the spatial arrangement of HCs is crucial for their function. Since all kinds of dyneins are attached to the same microtubule in the axoneme, there must be a tight coordination between different types of HCs in vivo. Variable inter-doublet spacing as proposed by Lindemann, or the relative orientation between adjacent microtubules may be important for the regulation of various types of dyneins.

1SAA-06 Shifting Gears with the Geometric Clutch

Charles B. Lindemann (*Oakland University, Department of Biological Sciences*)

When the axoneme of a cilium is bent, mechanical stress generates forces transverse to the outer doublets (t-forces). These t-forces act to push some doublets closer together or pry them apart. The geometric clutch (GC) hypothesis asserts that changes in the inter-doublet spacing caused by t-forces are responsible for the regulating the dynein motors to create the beat cycle. Computer models utilizing the GC mechanism have accurately simulated ciliary beating and a number of experimental results. However, based on an overview of recent evidence it may be necessary to revise at least one element of the original GC hypothesis. Specifically, dynein deactivation by t-force may occur while the dyneins remain attached to the B-subtubule of the adjacent doublet.

1SBA-01 Raman analysis of live cells with and without chemical markers

Shin-ichi Morita (*Tohoku Univ.*)

The use of Raman-tagged small molecules is the promising approach to purify our scientific interest. We are developing Raman tags, for instance, utilizing the resonance Raman effect to enhance the brightness of molecular emissions. We are also interested in developing Raman markers sensitive to surrounding environments.
Another promising approach is to use advanced spectral analysis to extract wanting information buried in the ambiguous and complicated data. Mainly in order to monitor dynamics of cellular differentiation, proliferation, and apoptosis, using a standard Raman microscope, it was possible to define cellular states, where the information of cellular conditions was successfully extracted. In the talk, the up-dated results will be introduced.

1SBA-02 １細胞解析のための発光センサーと光制御法の開発
Luminescent sensors and optical switches for single cell analysis

Takeaki Ozawa (*The University of Tokyo*)

A current focus of biological research is to quantify and image cellular processes in living cells. Herein, a new design of split FP and split luciferase will be described; the principle is based on reconstitution of the split-reporter fragments. To demonstrate the usefulness of the reconstitution technology, I will show some applications to imaging dynamics of endogenous RNA in single living cells. We used split-luciferase sensors for GPCR-β-arrestin interactions in living subjects. The concept of split-reporter reconstitution was further applied for a novel technology to manipulate kinase activities in living cells using external blue light. Herein, we will focus on recent advances in the imaging and manipulating biomolecules for single cell analysis.

1SBA-03 ラマン分光法を用いたマウス受精卵の分析
Analysis of mouse embryo by Raman spectroscopy

Mika Ishigaki[1], Kosuke Hashimoto[2], Kana Morimoto[1], Naoya Ogawa[2], Yukihiro Ozaki[1], Hidetoshi Sato[2] ([1]*Sci. and Tec. Chem., Kwan. Gak. Univ.*, [2]*Sci. and Tec. Biosci., Kwan. Gak. Univ.*)

The fertilized ovum contains all genetic information required to develop a complete organism. However, not all fertilized ova are viable. Some of them develop abnormally and ontogenesis stops. Various studies have mentioned that the survival rates of embryos are closely related with embryo quality. And the quality is generally assessed by blastomere morphology.
In our study, we non-invasively profile the molecular information about mouse embryo with Raman spectroscopy, and try to develop the new method to evaluate the embryonic quality based on molecular composition. We will discuss whether Raman spectroscopy can provide a new method for the non-invasive assessment and real-time monitoring of embryo quality or not.

1SBA-04 アルキン標識を用いた小分子のバイオラマンイメージング
Alkyne-tag Raman imaging of small molecules in biological systems

Jun Ando[1,2,3] ([1]*Dept. of Applied Physics, Osaka Univ.*, [2]*RIKEN*, [3]*AMED-CREST, AMED*)

Raman scattering microscopy has been utilized for visualizing molecular distribution and dynamics of biological systems. Recently, we have proposed to use alkyne as a Raman tag for imaging small molecules incorporated into living cells. Alkyne shows a distinct peak at Raman silent region of biomolecules, while it rarely changes the property of tagged parent molecule due to its tiny chemical structure compared with fluorescent labels. Alkyne-tagged thymidine analogue, coenzyme Q analogue, and mitochondria-selective probe have been visualized in cells by Raman microscopy. Here, I will review these topics on alkyne-tag Raman imaging, and present recent research progress on visualization of alkyne-tagged sphingomyelin in lipid rafts of an artificial monolayer membrane.

1SBA-05 ラマンイメージングのための化学的に活性化可能なアルキンタグの開発
Chemically-activatable alkyne tag for Raman imaging

Satoshi Yamaguchi (*RCAST, The Univ. of Tokyo*)

Alkyne-tag Raman imaging is a promising tool for non-invasive bioimaging. Different from fluorescent-tags, alkyne-tags are not bleached and smaller, and therefore, have advantages in visualizing the biomolecules of interest for a long period without major impact on their intrinsic properties. Recently, the stimuli-responsive ‘activatable’ fluorescent tags have been reported to clearly visualize the dynamic distribution changes even in high background signal. But so far, no activatable Raman-tag has been reported. Here, we first report a chemically-activatable alkyne-tag. In this study, the chemical activation of the alkyne-tagged cholesterol analog on lipid bilayer membranes was detected by Raman microscopy to show the potential as an activatable Raman probe.

1SCA-01 原子分解能ホログラフィーによる生体分子活性サイトの３D 原子構造直接解プロジェクト
Direct 3D atomic structure analysis project for active-site of bio-molecules by atomic-resolution holography

Hiroshi Daimon (*Nara Inst. Sci. Tech. (NAIST)*)

An active-site atom plays an important role in bio molecules such as the photochemical center of photosynthesis proteins. However the 3D atomic structure analysis around this center has been difficult by a standard structure analysis method such as an x-ray diffraction. Here several new holographic methods to analyze the 3D atomic structure around this kind of active center atoms are introduced such as “fluorescent x-ray holography”, “photoelectron holography” and “stereo photograph of atomic arrangement”. Recently, their accuracies to reproduce the atomic positions have improved dramatically. Hence we started a project of “3D active-site science” supported by JSPS Grant-in-Aid for Scientific Research on Innovative Areas: Grant Number 26105001.

1SCA-02 量子ビームを用いた 1 分子内部動態と活性サイト機能解析
Single Molecule Observations by Quantum Beams and Analysis of Functional Active-sites

Yuji Sasaki (*Department of Advanced Materials Science, Graduate School of Frontier Sciences, The University of Tokyo*)

We have proposed that single molecule techniques using X-rays, electrons, and neutron. Especially, Diffracted X-Ray Tracking has been developed for obtaining the information of the 3D internal motions of single protein molecules. This concept can apply to utilize by using both electrons and neutron. For example, Instead of the Laue diffraction using white X-ray, the Electron Back-Scattered Diffraction Pattern was adopted to monitor the 3D orientations of the nanocrystals linked to the single protein molecules. We called Diffracted Electron Tracking. Additionally, we call Diffracted Neutron Tracking for new single molecule measuring method in which the long time observation from the non-destructivity of a neutron is possible.

1SCA-03 アセチルコリン受容体のリガンド依存的チャネル開閉機構の解明に向けて ～X 線一分子計測と原子分解能ホログラフィーの可能性～
Toward understanding ligand-gated ion channels -Potential of diffracted X-ray tracking and atomic resolution holography-

Yuri Nishino[1], Hiroshi Sekiguchi[2], Yuji C. Sasaki[3], Atsuo Miyazawa[1] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*JASRI/SPring-8*, [3]*Grad. Sch. Frontier Sci., Univ. Tokyo*)

Nicotinic acetylcholine receptor (nAChR) is a ligand-gated ion channel and has at least three functional states (resting, active and desensitized states) depend on ligand-binding and channel gating. In order to understand the channel-gating mechanism of nAChR, combinational studies of atomic structure and molecular dynamics among each state should be informative. We observed the dynamic three-dimensional single molecule behavior of nAChR using a single molecule tracking technique, diffracted X-ray tracking (DXT) with pico-meter accuracy and micro-second time resolution. Dynamic information of nAChR's resting, active and desensitized states based on structure analysis will be discussed.

1SCA-04 ニトリル水和酵素の触媒機構に関する理論的研究
A QM/MM study of catalytic mechanism of nitrile hydratase

Megumi Kayanuma[1], Mitsuo Shoji[1,2], Yasuteru Shigeta[1,2] ([1]*Center of Comp. Sci., Univ. Tsukuba*, [2]*Grad. Sch. of Pure and App. Sci., Univ. Tsukuba*)

Nitrile hydratase (NHase), which catalyzes hydration of nitriles to the corresponding amides, is one of biocatalysts used in industrial productions. It has a unique active site structure which contains an Fe(III) or a Co(III) ion coordinated with two carboxamido nitrogens and three cysteine sulfer with different oxidation states (Cys-S, Cys-SO, and Cys-SO2). Several reaction paths have been proposed for the catalytic mechanism of NHase, and quantum chemical calculations using active-site models were reported. In the present study, we examine four reaction paths for initial steps of catalytic mechanism of NHase by using Quantum Mechanics/Molecular Mechanics (QM/MM) method and reveal the effects of surrounding protein environment on the reaction process.

1SCA-05 生体試料に向けた蛍光 X 線ホログラフィーの挑戦
Challenge of X-ray fluorescence holography toward biomaterials

Koichi Hayashi (*Institute for Materials Research, Tohoku University*)

X-ray fluorescence holography is a structural analysis method, which produce three dimensional atomic images around specific elements. To date, it has been applied to local structure analyses around dopants in inorganic materials. On the other hand, biological matters, such as hemoglobin, include metal atoms like dopants. Using X-ray fluorescence holography, we can visualize local structure around metal atoms. Thus, we started to apply the X-ray fluorescence holography to hemoglobin crystals. I will show some difficulties for achieving this task.

1SCA-06 ヘモグロビンのリガンド光解離過程における中間構造ダイナミクス観測
Structural dynamics measurements of the intermediate states in the ligand-photolysis of hemoglobin

Ayana Sato (*Sch. Med., Univ. Jichi*)

Hemoglobin (Hb) has tetrameric structure which consists of two pairs of αβ subunit with four protoheme, and can switch quaternary structure from T (tense) to R (relaxed) state caused by ligand (O_2, CO etc.) binding. Although Hb was one of the first protein structures ever to be solved by X-ray crystallography, its static structures do not tell us much about the gas migration pathway from the outside of the protein to the deeply buried heme. We report a series of X-ray crystal structures of the photolysis intermediates of COHb at 140 K, visualizing the time-dependent position of CO in the α and β subunits. We assign the individual ligand migration pathways in both subunits of Hb.

1SCA-07 ヘムの構造歪みの電子構造への影響に関する理論的研究
Theoretical study of the relationship between heme distortion and redox potential

Yu Takano[1,2], Yasuhiro Imada[2] ([1]*Grad. Sch. Info. Sci., Hiroshima City Univ.*, [2]*IPR, Osaka Univ.*)

Heme consists of an iron ion bound to the four central nitrogen atoms of a porphyrin ring. The iron-containing heme group is ubiquitous in biology. Heme proteins are involved in many biological roles such as electron transfer, oxygen transport, and catalysis. Protein environment affects the chemical properties of hemes for diverse functions of heme proteins. In this study, we have investigated the relationship between the degree of heme distortion and redox potential with density functional theory and a normal-coordinate structural decomposition procedure. Our computational data show that in-plane distortions of porphyrin ring contribute the change in redox potentials of hemes rather than out-of-plane distortion.

1SDA-01 マルチオミクスデータからのインスリン作用の大規模トランスオミクスネットワークの再構築

Reconstruction of a large-scale transomic network of acute insulin action from multi-omics data

Shinya Kuroda, Katsuyuki Yugi (*Biological Sciences, University of Tokyo*)

Cellular responses are composed of dynamic molecular interactions between multiple layers including protein phosphorylation, and metabolites. To reveal an unbiased whole picture, simultaneous quantitative and global measurements in these layers, rather than pin-point analysis of some selected molecules, is needed. Here, we simultaneously performed metabolomic and phospho-proteomic analysis in insulin-stimulated Fao hepatoma cells in collaboration with Prof. Soga (Keio Univ), and Dr. Matsumoto and Prof. Nakayama (Kyushu Univ), respectively, and developed an reconstruction method of insulin-dependent metabolic control pathway directly from trans-OMICS data. We demonstrate the global landscape of a transomic network of acute insulin action.

1SDA-02 Trans-Omics analysis of the central carbon metabolism in Saccharomyces cerevisiae

Fumio Matsuda, Hiroshi Shimizu (*Grad. Sch. Inform. Sci. Tech., Osaka Univ.*)

The central carbon metabolism is an old frontier of biology that should be revisited with trans-Omics data now. Whereas the central metabolic pathways in *Saccharomyces cerevisiae* as an model eukaryotic cell model have been characterized in 60s and 70s, a quantitative description of the complex behaviors has been attained by recent advances in the technologies for proteomics, metabolomics, and metabolic flux analysis that can produce a plentiful amounts of data. However, naive integration the data was hampered by an missing values of low abundance metabolites and enzymes, biological and analytical errors in the quantitative data, and poor theoretical background and kinetic model to precisely describe metabolic phenomena.

1SDA-03 タンパク質相互作用を利用した統計的全ゲノム相関解析

Statistical assessment for genome-wide association study with protein-protein interactions

Jun Sese (*BRD, AIST*)

Genome-wide association studies (GWAS) have been widely used for understanding the associations of SNPs with a disease. GWAS data are often combined with protein-protein interactions toward a system understanding of the biological changes caused by the SNPs. To determine which subgraphs are associated with the disease, a statistical test on each subgraph needs to be conducted. However, the lack of an analysis method causes no statistically significant results. We here introduce a method to enumerate subgraphs having statistically significant associations with a diagnosis. We then apply it to GWAS data from model species. This study might prioritize proteins whose structures should be investigated to understand diseases or drug targets.

1SDA-04 NF-κB pathway model の安定性・分岐解析

Stability and Bifurcation Analysis of an NF-κB pathway model

Gouhei Tanaka (*The University of Tokyo*)

The transcription factor NF-κB is mainly responsible for regulating the immune response to infection and associated with auto-immune diseases, cancer, and other disorders. It is significant to understand how NF-κB responds to various stimuli. The mechanism behind the regulation of NF-κB is still not fully understood, but the signal transduction pathways related to NF-κB have been found. In this study, we analyze a mathematical model of an NF-κB pathway. In particular, we focus on oscillatory phenomena and switch-like responses found in the model. We perform stability and bifurcation analysis of the model to clarify the condition for activation of the NF-κB. This work is in collaboration with K. Inoue, M. Okada, and K. Aihara.

1SDA-05 Dynamic behaviors of biochemical network

Mariko Okada (*RIKEN Center for Integrated Medical Sciences*)

Signaling-transcriptional networks are often controlled in a nonlinear manner. In antigen-stimulated B cell receptor (BCR) response, transcription factor NF-kappa B activity is controlled by positive feedback loops within the signaling pathway and negative feedback loops mediated by the transcriptional products, which result in a switch-like activation and oscillation behaviors of NF-kappa B. We mathematically modeled and experimentally analyzed a biochemical network of NF-kappa B. Our study shows that the characteristic dynamic behavior of NF-kappa B is truly modulated by the amount of particular proteins and regulatory loops in the network, thereby they controls expression of genes located at the downstream.

1SGA-01 スーパーコンピュータで明らかにするがんのヘテロ性

Unraveling cancer heterogeneity with supercomputer

Satoru Miyano (*The University of Tokyo*)

We present highly parallel software applications developed in the project "HPCI Strategic Programs for Innovative Research Field 1 "Supercomputational Life Science" (2011-2015) and the Grand Challenge Project for Life Science "Next-Generation Integrated Simulation of Living Matters (2006-2012). The first series of applications include various gene network estimation software applications and the second series include a series of software applications and pipelines for cancer genome analysis (exome, whole genome, RNA sequence). By using these software applications, we are currently making challenges to understand cancer heterogeneity on network-level from cancer big data.

1SGA-02 循環器系の階層統合シミュレーション
Hierarchical Integrated Simulations of Circulatory System

Shu Takagi (*The University of Tokyo*)

There are several stages of the simulations for cardiovascular system. We have been developed the software for entire vascular network model using the concept of 0D-1D-3D coupling. We have been also developing the multiscale thrombosis simulator. Recently, we have started the 3-D simulations for micro-circulation systems. So far, although these simulators are our own made, that is, developed with our colleagues, they are not unified for hierarchical integrated simulations of circulatory system. In the present talk, these simulators are briefly introduced and toward their hierarchical integration for the post "K-computer", future direction is discussed.

1SGA-03 個別化医療支援に向けたデータ同化生体力学シミュレーション
Biomechanical simulation integrated with clinical measurements toward personalized medicine

Shigeo Wada (*Graduate School of Engineering Science, Osaka University*)

The development of biomedical measurements such as MRI and X-ray CT has led to great progress in clinical medicine. On the other hand, recent advances in computational mechanics have enabled to simulate biological phenomena at various scales from cells to organs, bridging the distance between mechanics and biology. Thus, it is expected that the integration of the computational approach with the clinical data assists in individual diagnosis and treatment. However, the computational approaches have not been generally accepted in the clinical medicine where the biomedical measurement is emphasized as the evidence. This presentation describes the problems to be solved toward the personalized medicine.

1SGA-04 ポスト「京」重点課題 1：生体分子システムの機能制御による革新的創薬基盤の構築
Innovative drug discovery infrastructure through functional control of biomolecular systems by using post 'K' supercomputer

Yasushi Okuno[1,2] ([1]*RIKEN Quantitative Biology Center*, [2]*Graduate School of Medicine, Kyoto University*)

In the end of last year, developing of a next generation supercomputer, called post 'K' had been started. Following this project, we are also developing application and software for drug discovery and development by using post 'K'. The application project aims to develop ultra-high speed molecular dynamics simulations to achieve not only capturing long-time dynamics of target molecules in the time range of millisecond, but also controlling supra-molecular systems constituting of many biomolecules including factors that cause side-effects. We hope that post 'K' will accelerate discovery and design of safer and more highly effective drugs.

1SGA-05 Large-Scale Molecular Simulation of Viruses: Multi-scale Molecular Modeling Approach

Wataru Shinoda, Kazushi Fujimoto, Yoshimichi Andoh, Susumu Okazaki (*Grad. Sch. Eng., Nagoya Univ.*)

We illustrate here our recent work on the molecular modeling and simulations of large self-assembled macromolecular systems including virus capsid. Previous all-atom molecular dynamics simulations elucidated a special environment within a virus capsid, which motivated us a further investigation of molecular adsorption and entrance to the capsid in the context of drug design. To characterize the molecular mechanism, an efficient way to evaluate the free energy surface is desired. We here try a multi-scale molecular modeling approach to overcome this problem. We would like to discuss also a possible future work using high-performance supercomputers.

1SGA-06 多剤排出トランスポーター AcrB の動的構造変化
Conformational Dynamics of Multidrug Efflux Transporter AcrB

Mitsunori Ikeguchi (*Grad. Sch. Med. Life Sci., Yokohama City Univ.*)

Conformational dynamics of target proteins are crucially important in drug development especially for flexible proteins. Controlling the conformational dynamics of target proteins is one of major goals for HPC drug development using the post K supercomputer. In this talk, I present a computational study on conformational dynamics of the multidrug efflux transporter AcrB, which undergoes large conformational changes during drug transports. AcrB utilizes the concentration gradient of protons across the inner membrane of bacteria as an energy source. Coupling between two conformational changes in transmembrane and periplasm domains is responsible for dynamical functions of AcrB.

1SHA-01 クロマチンの構造多様性とダイナミクス
Structural versatility and dynamics of chromatin

Hitoshi Kurumizaka (*Faculty of Science and Engineering, Waseda University*)

In eukaryotes, genomic DNA is highly organized as chromatin, which functions as major regulatory factor for DNA metabolism. The basic unit, nucleosome, is composed of core histones, H2A, H2B, H3, and H4, and about 150 base pairs of DNA. In chromatin, nucleosomes are connected with linker DNA segments, and form the beads-on-a-strings structure, which is folded into the higher order chromatin architecture, probably with the chromatin associated proteins. We have studied the structure and physical character of the nucleosomes containing histone variants and modifications, and evaluated their contributions in higher order chromatin. In this symposium, current our progress on structural analyses of the various nucleosomes and poly-nucleosomes will be presented.

1SHA-02 X 線・中性子溶液散乱によるバリアント・ヌクレオソームの構造研究

Structural Investigation on Variant Nucleosomes by Solution X-ray and Neutron Scattering

Masaaki Sugiyama[1], Naoki Horikoshi[2], Yasuhiro Arimura[2], Rintaro Inoue[1], Akihisa Osakabe[2], Hitoshi Kurumizaka[2] ([1]*KURRI*, [2]*Dept. Elect. Eng. & BioSci., Waseda Univ.*)

Solution scattering by small-angle X-ray and/or neutron scatterings (SAXS/SANS) gives us structural information of solute. In the case of biomolecules, it is very helpful to understand the structures similar to those in vivo, such as structures fluctuated, modulated, not-crystalized and so on. SAXS is useful to observe the whole profile of protein whereas SANS can observe the partial structure utilizing its unique contrast variation technique. Therefore, by the complementary use, we can see the hidden structure. Following this concept, we analyzed the structures of variant nucleosomes and estimated how they are different from canonical one. In this presentation, we will report our recent results about H2A.B, H2A.Z nucleosomes.

1SHA-03 細胞核スケールにおける転写活性分布: ゲノム動力学シミュレーションからのアプローチ

Nuclear-scale spatial distributions of transcriptional activities: an approach from a 3D dynamical simulation model of yeast genome

Naoko Tokuda, Shin Fujishiro, Masaki Sasai (*Nagoya Univ.*)

To investigate the relationship between the nuclear-scale chromosomal conformation and gene expression, we have developed a 3-dimensional dynamical simulation model of genome of interphase budding yeast by using Hi-C data (Tokuda et al., Biophys J., 2012). Each coarse-grained chromosome was modeled by a kinkable fiber, and interactions among chromosomes were described by the Go-like potentials, which stabilize the intra- and inter- chromosomal distances at around their average values derived from the Hi-C data. With this simulation model, we investigated the reasons for the experimentally observed misregulation of 60 genes in a yku70 esc1 mutant (Taddei et al. Genome Res., 2009) by examining the mutational modification of spatial distribution of genes in nucleus.

1SHA-04 人工触媒システムによる『合成』エピジェネティクスを目指して

Toward synthetic epigenetics by artificial catalyst systems

Shigehiro Kawashima[1,2], Yoshifumi Amamoto[1,2], Hiroki Suto[1,2], Yuki Aoi[1,2], Nozomu Nagashima[1], Akihisa Osakabe[3], Yasuhiro Arimura[3], Hitoshi Kurumizaka[3], Kenzo Yamatsugu[1,2], Motomu Kanai[1,2] ([1]*Graduate School of Pharmaceutical Sciences The University of Tokyo*, [2]*ERATO Kanai Life-Science Catalysis Project*, [3]*Graduate School of Advanced Science and Engineering, Waseda University*)

Chemical modification of histones is crucial for epigenetic mechanisms that are involved in diverse biological processes and diseases. Here we report a novel artificial catalyst system to synthetically modulate epigenetics without relying on enzymes. We developed a novel catalyst, which catalyzes an acyl transfer from acyl-CoA, an endogenous acyl donor. By using suitable ligands for histones, the catalysts enabled selective introduction of acyl groups into specific lysine residues on histones. Therefore, artificial catalysts could substitute for endogenous HAT enzymes and control biological systems by synthetically modulating epigenetics in living cells. In addition, our catalyst system can be a unique tool to study the role of yet unexplored histone acylations.

1SHA-05 植物のクロマチン動態メカニズムに迫る

Studies of dynamic chromatin in plants

Sachihiro Matsunaga[1,2], Takeshi Hirakawa[1], Yuki Sakamoto[1], Takuya Sakamoto[1] ([1]*Dept. Appl. Biol. Sci., Fac. Sci. Tech., Tokyo Univ. Sci.*, [2]*JST, CREST*)

Nuclei have dynamically intranuclear structures with high integrity and plasticity. Recently increased knowledge of the dynamics of plant chromatin has revealed plant-specific robustness and participation in environmental responses. We investigated interphase chromatin dynamics of *Arabidopsis thaliana* by live cell imaging with a chromatin fluorescence tagging system, lacO/GFP-LacI system, which allows tracking of specific gene loci. We analyzed the DNA double strand break (DSB) response using the lacO/GFP-LacI system. We found that DSBs induced the approach of the homologous loci with γ-irradiation. Our screening of mutants for epigenetic regulators revealed that a chromatin remodelling factor, AtRAD54, was involved in chromatin dynamics with γ-irradiation.

1SHA-06 クロマチン修飾と転写活性化の in vivo ダイナミクス

Chromatin modification and transcription activation in vivo

Hiroshi Kimura (*Graduate School of Bioscience and Biotechnology, Tokyo Institute of Technology*)

Transcription is regulated in vivo in chromatin context, including the variation of nucleosome positioning, histone modification, and three-dimensional organization in the nucleus. As these factors exhibit dynamic changes in living cells, it is important to track functional and specific chromatin as well as transcription activity. By using Fab-based live endogenous modification labeling (FabLEM) technique, we have visualized histone modifications in different model systems for transcription activation. In all cases so far analyzed, histone H3 acetylation, rather than methylation, was most relevant to transcription activation. I will discuss the function of H3 acetylation on transcription kinetics in vivo.

1SIA-01 バイオミメティクス：生物多様性による技術革新

Biomimetics: Innovation Based on Biodiversity

Masatsugu Shimomura (*Chitose Institute of Science and Technology*)

Biological diversity is the result of a long evolutionary process for adaptation to various environments. This diversity has been acquired by “biological processes and techniques” including “production processes”, “operation principles” and “operation systems”, all of which differ from “human technology”. An interdisciplinary project based on the study of natural history, biology, agriculture, materials science, mechanical engineering, and environmental science, will open the door to a new system through the combination of lessons learnt from both “biological diversity” and “human wisdom”. A “biomimetics database” and information science is an important “device” of connecting biology and technology.

1SIA-02 昆虫のふしぎなつくり―驚異の構造と機能
A review of surprising structures and functions of insects

Shuhei Nomura (*Dept. Zool., Nat. Mus. Nat. Sci., Tokyo*)

The class Insecta (=insects) is a big group of arthropoda. It includes more than a million of taxonomical species, which occupies about three quarters of Animal Kingdom. The great biodiversity of insects is reached into various kind of habitats on the terrestrial parts of the world. In this presenrtation, camouflage and structural color of some insects are reviewed, and the associated microstructures of cicads, dragonflies, butterflies, etc. are explained together with SEM images. The presenter's project of database building is also introduced.

1SIA-03 海綿動物に学ぶバイオミメティクスの可能性
Sponges as potential model for biomimetics

Remi Tsubaki (*JAMSTEC*)

Sponges are sessile aquatic animals. They continuously flow ambient water through a vast canal system inside their body, and filter out and feed suspended organic particles. As sponges have no specialized organ such as digestive tract, they not only feed, but also breathe and breed by water flowing through the canal system. Their dependence on the canal system strongly suggests that the sponge canal system is likely optimized for efficient water transportation, which would potentially serve as an ideal model system for designing a water transportation system with high energy efficiency.

1SIA-04 生物の構造色とその光学効果
Structural color of animals and its optical effects

Shinya Yoshioka (*Tokyo Univ. of Sci.*)

It is well known that many species of animals utilize submicron structures to produce brilliant colors through optical phenomena such as interference, diffraction and scattering. These colors are called structural colors and many attempts have been performed to apply them in artificial products such as painting, textile, and cosmetics, because the structural colors have several useful optical properties, higher reflectance, spectral purity, and anti-fading property. I will talk about several examples of structural colors in nature. It will be shown that they have some additional mechanisms to add optical effects to the structural colors that are produced by periodic microstructures. Such additional mechanisms may be useful for the next-generation biomimetic engineering.

1SIA-05 ヤモリ模擬粘着剤の力学
Mechanics of Gecko Inspired Adhesives

Tetsuo Yamaguchi, Akira Akamine, Yoshinori Sawae (*Dept. Mech. Eng., Kyushu Univ.*)

Gecko has attracted much attention of a wide range of scientists and engineers due to its superior adhesion properties of its feet, i.e., they can attach with sufficient strength as gecko needs and can easily detach from an object so that it can walk not only on the ground, on the wall, but also on the ceiling. In this presentation, we report on the debonding process of a single gecko-inspired adhesive pad. We show that easy switching is possible with the structure. We also discuss the mechanics with a simple theoretical argument.

1SIA-06 生物に見られる微細・複合構造の生み出す材料物性：濡れ性と破壊靭性の観点から
Wetting and toughness of fine and composite structures in biological materials

Ko Okumura (*Ochanomizu Univ.*)

In nature, we find various fine and composite structures in biological materials. For example, on the surface of a leg of a certain roach, micron-scale blades are regularly aligned. This remarkable fine structure allows water intake necessary for the animal. In the case of lobsters, the exoskeleton is composed of three layers and two of them are composed of fiber bundles embedded in a matrix while the bundles form a spiral structure with a well-defined pitch. This surprising structure is the origin of the toughness of the exoskeleton. In this talk, we discuss simple physical mechanism for the wetting property and the toughness of the above biological materials, with some additional examples.

1SJA-01 アクチン線維のゆらぎはコフィリンのアクチン結合を制御する
Fluctuations of actin filaments modulate the binding of cofilin to the filament

Kimihide Hayakawa[1], Masahiro Sokabe[1], Hitoshi Tatsumi[2] ([1]*Mechanobiology, Nagoya Univ.*, [2]*Dept. of Appl. Biosci., Kanazawa Inst. of Tech.*)

Advances in the single-molecule manipulation and imaging have enabled an investigation of non-channel type cell mechanosensors, talin and p130Cas, focal adhesion proteins. Recently, actin filaments have been demonstrated to act as a "negative mechanosensor" in the presence of the actin-severing protein, cofilin. The cofilin severing activity negatively depends on the tension in the actin filament through tension-dependent binding of cofilin to the actin filament, which leads to that relaxed actin filaments are severed, while tensed ones are either not severed or severed after a long delay. Here we review the latest progress in the mechanosensing by actin filaments, and introduce the possible role of the fluctuations of the filament.

1SJA-02 アクチンフィラメントの協同的構造変化は、アクチン結合タンパク質との相互作用をアロステリックに制御する

Cooperative conformational changes of actin filaments allosterically regulate interactions with actin binding proteins

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Diverse functions of actin filaments are dependent on interactions with specific actin binding proteins (ABPs). Local biochemical regulation of each ABP has been implicated in localizing those interactions, but polymorphism of actin filaments is emerging as a novel mechanism of functional regulation of actin filaments. We present evidence to show that mutually exclusive cooperative binding of S1 (motor domain of myosin II) and cofilin to actin filaments is mediated by cooperative conformational changes of the filaments. High-speed AFM suggested that S1-mediated inhibition of cofilin binding involves highly cooperative untwisting of the filament structure. Similar allosteric regulation of ABPs may be widespread since many ABPs are known to change the filament structure.

1SJA-03 中性子準弾性散乱による F-アクチン、ミオシン S1 及び水和水のダイナミクス解析

Dynamics of F-actin, myosin subfragment-1 (S1), and their hydration water studied by quasielastic neutron scattering

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The picosecond dynamics of F-actin, myosin S1, and their hydration water were studied by quasielastic neutron scattering (QENS) at J-PARC. Analysis of the QENS spectra showed that a larger fraction of the atoms of F-actin undergoes the motions with the smaller residence time than S1. It was also found that the mobility of the hydration water of S1, which was evaluated from the translational diffusion coefficient and the residence time, is lower than that of bulk water, while that of the hydration water of F-actin is close to that of bulk water. These results suggest that the concerted action of rapidly fluctuating F-actin and its hydration water allows F-actin to explore a wide range of the conformational space, which would facilitate the binding of myosin to F-actin.

1SJA-04 アクチン繊維の「状態」とアクチン結合タンパク質

States of an actin filament associated with binding of other proteins

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Actin filaments are known to take various configurations in associated with polymerization or biding of other actin binding proteins. In order to know dynamic properties of the filament, it should be necessary to detect the states in solution. We have prepared the filament with localized portion which contains several tens of donor-acceptor pairs causing FRET. FRET efficiencies obtained from such portion of filaments, called "madara FRET filaments", were found to be classified into about 4 types, suggesting multiple states of actin filaments. Binding of myosin lowered the FRET efficiency of some classes and tropomyosin or dreblin increased. This technique may be powerful to reveal the fluctuations of actin filaments.

1SKA-01 V-ATPase のイオン透過機構

Ion transporting mechanism of V-ATPase

Takeshi Murata[1,2] ([1]*Science/Chiba-U*, [2]*PRESTO/JST*)

V-ATPases are ATP driven rotary proton pumps and are attractive drug targets for osteoporosis and cancer. The prokaryotic V-ATPase of Enterococcus hirae, closely related to the eukaryotic enzymes, provides a unique opportunity to study the ion-translocation mechanism because it transports Na+, which can be detected by radioisotope (22Na+) experiments and X-ray crystallography. We have obtained the biochemical data using the isotope, and the crystal structures of the membrane ring with Na+, Li+, or DCCD. The biophysical data of the membrane ring was also examined by ATR-FTIR spectroscopy and mass spectrometry in vacuum. In my talk, I would like to discuss about ion transporting mechanism of the V-ATPase based on these structural and biochemical, biophysical findings.

1SKA-02 電位依存性プロトンチャネルを介したプロトン透過経路の構造学的知見

Structural insight into proton conduction pathway via voltage-gated proton channel

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Voltage-gated proton channel, VSOP (or Hv1), consists of four transmembrane helices as a voltage-sensor domain. VSOP belongs to superfamily of voltage-gated ion channels, such as Kv or Nav, but lacks an authentic pore domain. In this study, we determined the crystal structure of mouse VSOP chimeric channel, which was optimized for crystallization. This structure showed unique features; not only resting state as a voltage sensor but also closed-state as a proton channel. Interestingly, two hydrophobic layers were found inside the VSOP protomer. These layers may prevent the penetration of water molecules as a proton transfer carrier. This double hydrophobic layer of VSOP may play distinct roles in proton pathway through plasma membrane.

1SKA-03 バクテリアのべん毛運動におけるイオン透過とエネルギー変換

Ion flax and energy transduction in bacterial flagellar motilty

Michio Homma (*Nagoya Univ.*)

Many bacteria have the flagellum for moving. The flagellum has motor embedded in membrane and is necessary to generate torque by flowing coupling ions, H^+ or Na^+, by electrochemical potential of cells. The motor has driven by the interaction between two components, the stator and the rotor. The stator complex, which is composed of MotA/MotB for H^+-driven motors and PomA/PomB for Na^+-driven motors. The A and B subunit is a protein with 4-transmembrane (TM) domains and with a single TM domain, respectively. The B subunit has an essential charged residue in the TM region and a plug region which is opened by a conformational change. In this talk, I want to discuss the ion flax and the energy transduction in the H^+ - or Na^+-driven type motor.

1SKA-04 プロトンポンプは何を運んでいるのか？
What do H^+ pumps transport?

Hideki Kandori (*Nagoya Inst. Tech.*)

Na^+ and Cl^- pumps transport Na^+ and Cl^-, respectively. Then, what about H^+ pump? It is well established that bacteriorhodopsin (BR) is a light-driven outward H^+ pump. However, about 15 years ago, an interesting proposal was reported that BR is not an outward H^+ pump, but an inward OH^- pump. This proposal has never been denied for BR. This fact demonstrates unique aspect of H^+ transport, being in clear contrast to the transport of other ions. H^+ pump is the result of H_3O^+ transport, OH^- transport, or Grotthuss mechanism (conserted H^+ transfer through water chain). Grotthuss mechanism seems to be dominant for H^+ pumps and channels, but it is not easy to give an experimental evidence. I will summarize recent understanding on light-driven H^+ pumps in my talk.

1SKA-05 分子動力学シミュレーションを用いた電位依存性プロトンチャネル VSOP における亜鉛イオンの影響の解析
Molecular dynamics simulations of the effect of zinc on the voltage-gated proton channel VSOP

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Voltage-gated proton channel VSOP, which contains four-transmembrane segments homologous to the voltage-sensing domain of conventional voltage-gated ion channels, is activated at depolarized voltage and sensitive to pH gradient and binding of zinc ions. The double mutations of the two acidic residues (E115 and D119) or the histidine residues (H136 and H190) around the zinc-binding site make VSOP insensitive to zinc ions. In order to analyze molecular mechanisms of these regulations and effects of zinc ions we performed molecular dynamics simulations of the wild-type and mutant proteins (E115S, D119S, and E115SD119S). These results suggest that either E115 or D119 is required for the stabilization of a zinc ion.

1SKA-06 チトクロム c 酸化酵素の酸化還元反応と共役したプロトン取り込み・排出機構
Molecular mechanism of redox coupled proton uptake and release processes of Cytochrome c oxidase

Yasuteru Shigeta[1], Katsumasa Kamiya[2] ([1]*Department of Pure and Applied Sciences, University of Tsukuba*, [2]*Center for Basic Education and Integrated Learning, Kanagawa Institute of Technology*)

Cytochrome c oxidase (CcO) is the terminal oxidase of cellular respiration. It reduces dioxygen to water, and this reaction is coupled with proton-pump activity. The CcO X-ray structure investigations have provided with high precision the information on the relative positions of their amino acid residues. However, an a posteriori analysis of the X-ray structures is occasionally needed to reveal the complex mechanisms of the underlying chemical reactions. We here report first-principles calculations based on QM/MM performed on the entrance of D pathway and exit of H pathway in bovine CcO. Our results suggest the redox-controlled proton-transfer-coupled rotations at these sites are key processes for preventing a back flow and an excess influx of protons from bulk.

1SAP-01 生細胞内における温度計測と操作による温度生物学
Imaging and manipulation of intracellular temperature for thermal biology

Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

Temperature, a key regulator of biochemical reactions, influences many physiological functions of organisms. Recent progress in intracellular thermometry shows temporal and spatial variation associated with cellular functions, shedding light on an intriguing hypothesis: temperature change inside of a cell is essentially involved in cell biology. Conceiving this idea, we have been investigating intracellular temperature based on a fluorescent polymeric thermometer and quantitative fluorescence imaging. Furthermore, we manipulated intracellular temperature by irradiating infrared laser onto a cell to observe the thermodynamics and cell response. These techniques are indispensable for the advancement of thermal biology, which explores the commitment of temperature to life.

1SAP-02 ASK1 signaling regulates brown adipocyte maturation

Kazuki Hattori, Hidenori Ichijo (*Laboratory of Cell Signaling, Graduate School of Pharmaceutical Sciences, The University of Tokyo*)

Brown adipose tissue is in the spotlight as a therapeutic target for some metabolic diseases. ASK1, a member of MAP kinase cascade, became evident that it is highly expressed in adipose tissues; however the function of ASK1 remained to be identified. Through DNA microarray analysis, we revealed that the expression levels of specific subset of genes, which are known to be involved in thermogenesis, were suppressed in ASK1-deficient brown adipose tissue. Further analyses using primary culture of brown adipocytes elucidated that PKA-ASK1-p38 pathway regulates gene induction during the course of differentiation. The fact that ASK1-deficiency led to impaired metabolic responsiveness shows the significance of ASK1 in the function of brown adipose tissue.

1SAP-03 TRP チャネルによる温度感知の分子機構
Moleculae Mechanisms of Thermosensation through TRP channels

Makoto Tominaga[1,2] ([1]*Okazaki Institute for Integrative Bioscience*, [2]*SOKENDAI*)

TRP (transient receptor potential) channels comprise six related protein families (TRPC, TRPV, TRPM, TRPA, TRPML, TRPP). One subunit of the TRP channel is composed of six transmembrane domains and a putative pore region. Some TRP channels have been proven to be involved in thermosensation detecting ambient temperatures from cold to hot. There are now ten thermosensitive TRP channels (TRPV1, TRPV2, TRPV3, TRPV4, TRPM2, TRPM3, TRPM4, TRPM5, TRPM8 and TRPA1) with distinct temperature thresholds for their activation. I will to talk about the structure and physiological significance of several thermosensitive TRP channels. In addition, how structure and functions of thermosensitive TRP channels were changed dynamically in the process of evolution will also be discussed.

1SAP-04 体温の中枢神経調節
Central regulation of body temperature

Kazuhiro Nakamura[1,2] ([1]*Dept. Integrative Physiol., Nagoya Univ. Grad. Sch. Med.*, [2]*PRESTO, JST*)

Maintaining body temperature under a variety of thermal environments is one of essential homeostatic functions in homeothermic animals including humans. Body temperature is regulated by the central circuitry system through its thermosensory afferent mechanism and commanding efferent mechanism. The central thermoregulatory system is also employed to develop fever for the host defense against infection as well as to drive psychological stress-induced hyperthermia to survive the "fight or flight" situation under stress conditions. In this symposium, I will summarize recent knowledge of the central circuitry mechanisms for autonomic regulation of body temperature as well as for the development of fever and stress-induced hyperthermia.

1SAP-05 熱ストレスで駆動する核—細胞質間輸送を担う Hikeshi
Thermal stress-induced nucleocytoplasmic transport mediated by Hikeshi

Naoko Imamoto (*RIKEN*)

We recently showed that the system of nucleocytoplasmic transport is significantly affected by thermal stress. During thermal stress, transport pathways mediated by well known importin β family members are down regulated, whereas a novel transport pathway mediated by unique protein named Hikeshi is up regulated. Hikeshi mediates nuclear import of molecular chaperone Hsp70s. Hikeshi is evolutionary conserved protein present in most eukaryotic cells. Manipulation of Hikeshi proteins in different organisms shows that this protein is involved in glucose deprivation, senescence, and neurodegenerative disease. We aim to understand molecular basis underlying these phenomena in connection to the function of Hikeshi seen in cultured mammalian cells associated by thermal stress.

1SAP-06 局所熱パルス法による単一細胞の機能制御
Regulation of cellular functions by microscopic heat pulses

Kotaro Oyama[1,2], Madoka Suzuki[3,4], Shin'ichi Ishiwata[2,3,4] ([1]*Dept. Cell Physiol., The Jikei Univ. Sch. Med.*, [2]*Sch. Adv. Sci. Eng., Waseda Univ.*, [3]*WASEDA Biosci. Res. Inst. Singapore (WABIOS)*, [4]*Org. Univ. Res. Initiatives, Waseda Univ.*)

Local and temporary temperature changes in living cells are caused by chemical reactions within the cells and the environment. The changes have the potential to modulate the activity of both the cells themselves and also the surrounding cells. The viewpoint of "Thermo-Chemical Signaling" should be essential to understand how the living cells efficiently use the surrounding local temperature changes and those within the cell itself. In this viewpoint, we have investigated the biological roles of local temperature changes with a combination of microheating systems and fluorescent thermometers under the optical microscope. We would like to introduce the microheating-induced unique reactions of various single cells such as muscle contractions and neurite outgrowth.

1SAP-07 線虫 C. elegans における経験に依存した低温馴化機構
Temperature experience-dependent cold acclimation in nematode C. elegans

Tomoyo Ujisawa, Kohei Ohnishi, Tohru Miura, Akane Ohta, **Atsushi Kuhara** (*Institute for Integrative Neurobiology, Konan Univ.*)

Animals can acclimate to temperature. C. elegans has cold acclimation that is useful model to study temperature sensation and memory (1). 15 degree-cultivated animals can survive at 2 degree, while 20 degree-cultivated animals can not survive at 2 degree. We found that a pair of light sensoryneurons (ASJ) regulate cold acclimation. Calcium imaging determined that ASJ senses temperature. Common trimeric G-protein pathway is used between light and temperature signaling. Insulin released from ASJ is received by intestine and neurons, in which gene expression is controlled. We thus demonstrated novel system for cold acclimation (1). We are now screening GPCR-type temperature receptor and memory molecules in cold acclimation.

(1) Ohta, Ujisawa et al, Nature commun, 2014

1SBP-01 Target search process of a tumor suppressor p53 revealed by single-molecule fluorescence microscopy

Kiyoto Kamagata (*IMRAM, Tohoku Univ.*)

p53 can search for and bind to specific target sites of DNA, which results in cell cycle arrest, apoptosis and DNA repair. How does p53 search the target sites among a huge DNA within a physiological time? In this study, we developed a new method for making the array of aligned DNAs, DNA garden, and investigated the target search dynamics of p53 by using a single-molecule fluorescence microscopy. 1D sliding of p53 was strongly dependent on the concentration of Mg and Ca, and the sliding distance of p53 was maintained in loss of homeostatic control. We found two sliding modes which correspond to two binding forms of p53. The binding probability of p53 to the target site was regulated by the mutation and posttranslational modification.

1SBP-02 分子シミュレーションによる蛋白質-DNA 相互作用の構造可塑性
Flexibility in protein-DNA interactions studied by molecular simulations

Shoji Takada (*Dept. Biophys., Kyoto Univ.*)

Most DNA-binding proteins have multi-domains that include intrinsically disordered linkers and/or tails. Through such flexible nature, proteins can radically change their conformation depending of interaction with DNA. Moreover, via interaction with proteins, DNA also can change its structure. We have been studying these large-scale structural changes in proteins-DNA complexes using molecular simulation approaches. We report our recent studies on transcription factors, such as p53, binding to DNA and nucleosome systems.

1SBP-03 細胞機能の制御に向けた DNA 結合タンパク質の創製
Design of Artificial DNA Binding Proteins towards Manipulation of Cellular Functions

Miki Imanishi (*ICR, Kyoto Univ.*)

Artificial DNA binding proteins binding to desired DNA sequences are useful to edit genome sequences or control transcription of various genes. Using a C2H2-type zinc finger motif, we designed an artificial transcription factor that led to circadian phase resetting. We also manipulated gene expression patterns to be circadian by fusing a core clock protein with a zinc finger domain. Besides zinc finger motifs, Transcription Activator-Like Effectors (TALEs) have been used as templates for sequence specific artificial DNA binding proteins. However, almost all TALE binding sites are preceded by a 5' terminal T nucleotide. We performed directed evolution of TALEs to bind to any 5'-bases, which would expand possible target sequences.

1SBP-04 RNA 結合タンパク質のＲＮＡ界面予測方法の向上とその適用
Improvement in the prediction of RNA interface in RNA-binding Protein

Kei Yura[1,2] ([1]*Center for Info. Biol., Ochanomizu Univ.*, [2]*NIG*)

RNA-binding proteins together with RNA molecules play significant roles in the various aspects of cellular systems. Recent development of the new methods to identify global views of RNA-binding specificity further emphasizes the importance in molecular understanding protein-RNA interactions. We have developed KYG, a simple method to predict RNA-binding residues from the structure of RNA-binding proteins and the method has been used for a number of experiments performed by other groups. In this talk, we will introduce an improved method for detecting RNA interface residue by incorporating, for instance, a recursive calculation. With the increase of RNA-protein complex structures in PDB, we can improve the accuracy and widen the area of application of this method.

1SBP-05 蛋白質・RNA 複合体の立体構造予測
Three dimensional protein-RNA complex structure prediction

Tomoshi Kameda[1], Junichi Iwakiri[2], Michiaki Hamada[3], Kei Yura[4,5], Kiyoshi Asai[1,2] ([1]*Biotech. Research Inst. for Drug Discovery, AIST*, [2]*Grad. School of Frontier Sciences, Univ. of Tokyo*, [3]*Faculty of Science and Engineering, Waseda Univ.*, [4]*Grad. School of Humanities and Sciences, Univ. of Ochanomizu*, [5]*NIG*)

RNA-protein interactions play fundamental roles. To understand it, the three-dimensional (3D) structures of RNA-protein complexes are important. However, 3D structure determination of complexes is often difficult, suggesting that an accurate 3D structure prediction method is needed.

We propose two prediction methods: one is based on docking for predicting 3D structure of protein-protein complex. We modified it for RNA-protein complex, and the result shows its prediction accuracy is the highest level in the world.

The other is based on MD simulation and KYG algorithm. The advantage to previous method is input information: previous method requires the apo form structures of both protein and RNA, this method only does protein structure. Details will be talked in the session.

1SBP-06 リボアート・システムを用いたアプタマー創薬
Therapeutics Aptamer Discovery by RiboART System

Yoshikazu Nakamura (*UTokyo IMSUT RNA Medical Science*)

Aptamer is a folded single-stranded nucleic acid that binds given molecules. The concept is based on the ability of short (20-80 mer) sequences to fold, in the presence of a target, into unique three-dimensional structures, which allow the aptamer to bind target molecules with high affinity and specificity. We have been engaged in creating therapeutic RNA aptamers for unmet medical needs using the RiboART system for Ribomic Aptamer Refined Therapeutics. In this talk, several RIBOMIC programs will be presented from the discovery stage to the in vivo animal model study, including the X-ray structural study accomplished in collaboration with Prof. Osamu Nureki's group of UTokyo.

1SFP-01 タンパク質と脂質分子の協調
Cooperation among protein and lipid molecules

Yoshinori Fujiyoshi (*Grad. Sch. Pharma. Sci./CeSPI, Univ. Nagoya*)

Structure analysis of water channel AQP4 at 2.8 Å by electron crystallography shows 8 water densities, whereas the densities analyzed at 1.8 Å by X-ray crystallography were blurred. The counter intuitive notion could be attributed to different conditions of structure analyses of AQP4 molecules with lipid molecules or without. If a molecule acquired two important functions of channel and cell adhesion, the molecule could regulate physiologically complex functions. I therefore termed these molecules "adhennels" by concatenating two words, adhesive and channels. Both functions are accomplished by cooperation among membrane proteins and lipid molecules. We would like to discuss these multifunctional channels including gap and tight junction from structural point of view.

1SFP-02 生体膜におけるリン脂質分子の運動と機能
Probing the molecular motion and function of membrane phospholipids

Masato Umeda (*Dept. Syth.Chem and Biol.Chem. Sch. Eng. Kyoto Univ.*)

The importance of the specific lipid-protein interactions has been indicated by recent crystallization analyses of membrane proteins. What remains to be clarified is to understand the nature and biological function of dynamic lipid-protein interactions in cellular membranes. We have established a series of phospholipid-specific binding probes and explored the molecular motion and function of membrane phospholipids. We recently demonstrated that the transbilayer movements of membrane phospholipids, which are mediated by the P4-ATPase phospholipid flippase complex, play a crucial role in controlling cell migration and fusion. Molecular mechanisms underlying how the localized changes in phospholipid asymmetry affects the protein function will be discussed.

1SFP-03 Dynamics of Pore Forming Toxins during Their Assembling on Lipid Membranes

Neval Yilmaz, Toshihide Kobayashi (*RIKEN*)

Pore-forming toxins (PFTs) are soluble proteins that bind to membrane receptors and oligomerize to form pores. The molecular mechanism of assembling of PFTs and the interaction of PFTs with membranes are not well understood. High-speed atomic force microscopy (HS-AFM) enables the real-time visualization of the dynamics of PFT-membrane interaction at nanometer scale. Our HS-AFM results revealed the dynamic process of the assembling of sphingomyelin (SM)-binding PFTs on supported lipid bilayers. Together with cholesterol, SM forms specific lipid raft domains in biological membranes. HS-AFM results also suggest that PFT induces lipid-phase mixing in an oligomerization-dependent manner.

1SFP-04 Structure and Dynamics of Membrane Lipid Nanodomains using High-Speed AFM

Pierre-Emmanuel Milhiet (*CBS, CNRS/INSERM*)

Plasma membrane of eukaryotic cells are now described as a mosaic of micro or nanodomains that are highly dynamic and can segregate to form functional platforms. In order to better understand the molecular mechanisms associated to the formation of membrane domains through lipid lateral segregation, we analyzed partition of the ganglioside GM1 using artificial lipid bilayers presenting lipid phase separation. Using both standard and high-speed atomic force microscopy (HS-AFM), we demonstrated the propensity of GM1 to form stable nanodomains that partition into lipid-ordered phase. These domains were characterized in terms of shape, diffusion coefficient and motion mode using HS-AFM.

1SFP-05 High-Speed Atomic Force Microscopy: Watching Dynamic Processes at the Membrane at High Spatio-Temporal Resolution

Lorena Redondo, Atsushi Miyagi, Ignacio Casuso, **Simon Scheuring** (*INSERM/Aix-Marseille U.*)

The advent of high-speed atomic force microscopy (HS-AFM) has opened a novel research field for the dynamic analysis of single bio-molecules. The endosomal sorting complex required for transport (ESCRT) mediates membrane remodeling in cells. We used HS-AFM to study the ESCRT-III complex, i.e. Snf7. HS-AFM movies reveal Snf7 complex formation from filaments to maturated assemblies: Interfilament dynamics provide basis for a mechanistic understanding of tension generation for membrane fission. Annexin-V (A5) binds to negatively charged lipid bilayers in the presence of Ca2+ for membrane healing. Using a HS-AFM coupled to a buffer exchange flow system, we found two classes with different apparent affinity in the reversible association-dissociation of A5 to the membrane.

1SFP-06 脂質分子が膜タンパク質機能に及ぼす役割
Role of lipid molecules on molecular functions of membrane proteins

Yuji Sugita (*RIKEN Theoretical Molecular Science Laboratory*)

Many membrane proteins act as channels, pumps, and transporters in the biological membranes. Since they are surrounded by various lipid molecules, interaction between lipids and membrane proteins seems to be important. Recently, we have carried out all-atom molecular dynamics (MD) simulations of several membrane proteins, namely, SecY translocon, MATE multi-drug transporter, SR Ca-ATPase, Amyloid Precursor Protein (APP), and so on. In the MD trajectories, we observed specific protein-lipid interactions, some of which may be difficult to observed directly by experiments. In the talk, we discuss how the interaction enhances the conformational stability or to regulate the molecular functions.

1SGP-01 SACLA の現状と生物学研究実験
Current Status of SACLA and Experiments for Biological Studies

Kensuke Tono[1,2] ([1]*JASRI*, [2]*RIKEN SPring-8 Center*)

X-ray free electron lasers provide intense and coherent X-ray pulses with ultra-short duration in the femtosecond order. These properties have been stimulating the development of new measurement schemes in X-ray experiments such as X-ray diffraction, scattering, spectroscopy, and imaging for biological targets. One typical example is serial femtosecond crystallography (SFX), in which randomly-oriented crystals are dispersed in fluid and delivered to the interaction point with a fluid injector. Diffraction images are recorded in a shot-by-shot manner.
In this talk, we will give an overview of SPring-8 Angstrom Compact free-electron LAser (SACLA) and recent applications to biological studies, and introduce SFX instruments developed at SACLA.

1SGP-02 オルガネラの XFEL 低温コヒーレント X 線回折イメージング
Cryogenic coherent X-ray diffraction imaging of cellular organelle by using XFEL

Yuki Sekiguchi[1,2], Amane Kobayashi[1,2], Tomotaka Oroguchi[1,2], **Masayoshi Nakasako**[1,2], Yuki Takayama[2], Masaki Yamamoto[2], Yayoi Inui[3], Sachihiro Matsunaga[3], Yuichi Ichikawa[4], Hitoshi Kurumizaka[4], Mitsuhiro Shimizu[5] ([1]*Sci. Tech., Keio Univ*, [2]*RIKEN SPring-8 Center*, [3]*Sci. Tech., Tokyo Univ. Sci.*, [4]*Sci. Tech., Waseda Univ.*, [5]*Sci. Tech., Meisei Univ.*)

Coherent X-ray diffraction imaging (CXDI) is a technique visualizing the structures of non-crystalline particles with the dimensions of sub-micrometer to micrometer at resolutions of tens of nanometer. In CXDI experiments, specially isolated non-crystalline particles are irradiated by coherent X-rays. The electron density maps projected along the direction of X-ray are reconstructed by using the phase-retrieval algorithms. We have developed cryogenic X-ray diffraction apparatuses to collect a huge number of diffraction patterns of organelle particles using X-ray free electron laser. Here we report recent experimental results to reveal structural changes inside cellular organelle during cell cycles at resolutions better than 100 nm.

1SGP-03 X線レーザー回折による生細胞イメージング
Imaging live cells by X-ray laser diffraction

Yoshinori Nishino[1], Takashi Kimura[1], Yasumasa Joti[2], Yoshitaka Bessho[3] ([1]*RIES, Hokkaido Univ.*, [2]*JASRI/SPring-8*, [3]*IoP, Academia Sinica*)

Coherent imaging is a growing field in optical science. It requires no lens for image formation, but instead numerically reconstructs object images from the coherent diffraction data. Recently emerging X-ray free-electron lasers (XFELs) further extends the ability of coherent imaging to achieve spatial resolution beyond the conventional radiation-damage limitation. Thanks to the femtosecond pulse duration of XFEL, X-ray interaction with the sample occurs before radiation damage becomes obvious. We are developing a method which we refer to as pulsed coherent x-ray solution scattering (PCXSS) using the Japanese XFEL facility SACLA. PCXSS enables us to image living cells in solution at nanometer resolution.

1SGP-04 重原子誘導体を利用したシリアルフェムト秒結晶学による新規構造決定
De novo structure determination for serial femtosecond crystallography using heavy atom derivatives

Keitaro Yamashita[1], Dongqing Pan[2], Michihiro Sugahara[1], Hideo Ago[1], Masaki Yamamoto[1], Toru Nakatsu[1,2] ([1]*RIKEN/SPring-8*, [2]*Dep. Struct. Biol., Grad. Sch. Pharm. Sci., Kyoto Univ.*)

Serial femtosecond crystallography (SFX) with X-ray free electron lasers (XFELs) holds great potential for structure determination of challenging proteins that are not amenable to producing large well diffracting crystals. Efficient de novo phasing methods are highly demanding and as such most SFX structures have been determined by molecular replacement methods. We applied the single isomorphous replacement with anomalous scattering (SIRAS) method to SFX de novo phasing with a significantly reduced number of single-shot diffraction patterns. We employed high energy X-rays from SACLA to take advantage of the large anomalous enhancement near the absorption edge of Hg, which is one of the most widely used heavy atoms for phasing in conventional protein crystallography.

1SGP-05 SACLAにおける時分割シリアルフェムト秒結晶構造解析：バクテリオロドプシンの光反応中間体構造について
Structural dynamics of bacteriorhodopsin using time-resolved serial femtosecond crystallography at SACLA

Eriko Nango (*RIKEN RSC*)

Serial femtosecond crystallography (SFX) using ultrashort pulses from XFEL is an emerging method to enable structure determination without radiation damage. Protein microcrystals are carried by an injector serially and each diffraction pattern from crystals can be recorded before the onset of radiation damage. Furthermore, SFX holds promise for ultrafast time-resolved study of proteins. Laue diffraction or freeze-trapping methods is known as conventional time-resolved methods. However, the determination of time-resolved protein structures is hampered by various difficulties such as radiation damage. Recently, we have developed a new system for time-resolved SFX at SACLA. I will give a talk about structural dynamics of bacteriorhodopsin by using time-resolved SFX.

1SGP-06 巨大タンパク質の高分解能・無損傷結晶構造解析が可能なフェムト秒X線結晶構造解析法の開発
Development of a method of femtosecond crystallography enabling determination of high-resolution native structure of huge proteins

Hideo Ago[1], Kunio Hirata[1,2], Go Ueno[1], Masaki Yamamoto[1], Kyoko Shinzawa-Itoh[3], Tomitake Tsukihara[2,3,4], Shinya Yoshikawa[3], Michihiro Suga[5], Fusamichi Akita[5], Jian-Ren Shen[5] ([1]*RIKEN SPring-8 Center*, [2]*CREST, JST*, [3]*Grad. Sch. Sci., Univ. Hyogo*, [4]*Institute for Protein Research, Osaka Univ.*, [5]*Grad. Sch. Nat. Sci., Okayama Univ.*)

Precise structural information from high-resolution X-ray protein crystallography (PX) is widely used in molecular functional analysis. Due to possible X-ray induced radiation damage, however, the structure determined by PX may not always represent the native structure accurately.
We developed a new method of femtosecond crystallography that enabled native structure determination of huge proteins without radiation damage at high resolution by combined usage of XFEL pulses and crystals compatible with conventional PX in size. In this presentation I will introduce this new method and its application to two huge metalloenzymes highly susceptible to radiation damage [(1, 2)].
(1) Hirata, K. *et al.* (2014) *Nature Methods* **11**, 734-736.
(2) Suga, M. *et al.* (2015) *Nature* **517**, 99-103.

1SHP-01 X線繊維回折法による微小管内チューブリン構造の動態解析
Microtubule dynamics revealed by the X-ray fiber diffraction analysis

Shinji Kamimura[1], Hiroyuki Iwamoto[2] ([1]*Dept. Biol. Sci., Chuo Univ.*, [2]*JASRI, SPring-8*)

One of the most fundamental questions on the properties of microtubule is how its dynamics is related to the conformational changes of tubulin within the native microtubules. To address the issues, we applied our new technique for the quick shear-flow alignment of biological filaments, which enabled us to acquire fine X-ray fiber diffraction signals in a few seconds. We found that microtubules could be classified into three main groups with distinct axial periodicities of tubulin, which varied depending on GTP-hydrolysis and the contents of microtubule stabilizers, suggesting tubulin can undergo dynamic conversions among different states. After a short introduction to our novel fiber diffraction technique, discussions on the tubulin molecule dynamics will be given.

1SHP-02 The structure of kinesin bound to tubulin links the nucleotide cycle to movement

Benoit Gigant (*I2BC, CNRS, France*)

Kinesin-1 is a dimeric motor protein that moves along microtubules as it hydrolyses ATP. But the connection between the chemical energy used and the mechanical work produced has remained elusive, mostly because the high resolution structure of microtubule-bound kinesin was not known. We determined the structure of kinesin in the nucleotide-free and in an ATP-like states in complex with the microtubule track protein, tubulin, leading to the description of the kinesin structural cycle. The comparison of these structures shows in particular that conformational changes induced upon ATP binding may be described by rigid-body motions of three subdomains. These movements trigger a mechanical step and condition both the directionality and the processivity of the kinesin.

1SHP-03 微小管における構造柔軟性と、モーター蛋白の運動メカニズム

Conformational flexibility of microtubule linked to motility mechanism of motor proteins

Etsuko Muto (*RIKEN, Brain Science Institute*)

Our earlier study using in vitro motility assay showed that a kinesin-coated latex bead binds to a microtubule in a cooperative manner, indicating that microtubules, which have previously been considered to be passive tracks for kinesin, are actually dynamic. More recently, using mutational analysis of tubulin, we found that both kinesin and dynein use precisely the same loop/helix in alpha-tubulin for their motility and ATPase activation, despite the fact that these two motors belong to different enzyme classes. As the loop is a priori flexible, the structural pathway of ATPase activation should involve the conformational change of tubulin. Our findings open up a new perspective of the motor-microtubule interaction.

1SHP-04 Conformational switching of tubulin serves as the guidance cue for the intracellular transport by kinesin

Yasushi Okada (*Quantitative Biology Center, RIKEN*)

The structures of kinesin and the kinesin-tubulin complex have been solved by cryo-EM and X-ray crystallography, which deepened our understanding of the motility mechanism. Recent high resolution cryo-EM structures of microtubule also suggested conformational switching underlying the dynamic instability of microtubules. However, it is still unclear how the conformational switching of microtubules affects the binding of kinesin and its motility. We measured the affinity of kinesin to both GTP- and GDP- forms of microtubules in vitro and compared the results with the in single molecule live cell imaging. Based on these results, we would discuss on the new regulatory mechanism of kinesin by the conformational switching of tubulin in the microtubule lattice.

1SHP-05 Conformational switching of tubulin by Alp7/14 TOG-family polymerase

Frauke Hussmann, Douglas R Drummond, Daniel Peet, Douglas S Martin, **Robert A Cross** (*Warwick Medical School*)

Alp14, a TOG family microtubule polymerase from S. pombe, accelerates microtubule plus end growth. To probe its mechanism, we progressively reduced GTP-tubulin concentration whilst increasing the Alp14 concentration to maintain the plus end growth rate. This revealed that Alp14 can drive the plus end growth of S. pombe microtubules at GTP-tubulin concentrations considerably below the usual critical concentration for plus end growth. Alp14 is thus an unconventional enzyme that shifts the equilibrium constant for the reaction that it catalyses. We propose that the TOG domains of Alp14 recruit GTP-tubulin and switch it into a partially-bent conformation, whilst the Alp14 C-terminal tail regions independently bind to and hyperstabilise the GTP cap.

1SHP-06 ダイナミックな微小管細胞骨格は植物細胞の形を作り出し、環境ストレスに応答する

The dynamic microtubule cytoskeleton builds the shape of plant cells and responds to environmental stresses

Takashi Hashimoto (*Graduate School of Biological Sciences, Nara Institute of Science and Technology*)

Cortical microtubule (MT) arrays in the interphase plant cell spread in a quasi-2D sheet in association with the inner face of the plasma membrane, guiding the movement of cellulose-synthase complexes, and thereby controlling the cell's directional expansion and shape. Molecular genetic screens in Arabidopsis thaliana have identified various tubulin mutants and MT regulators that affect directional expansion of plant cells. We also found that acute hyperosmotic stresses activate novel tubulin kinase and induce rapid and transient generation of polymerization-inefficient tubulin isotypes, resulting in reorganization of cortical MT arrays. We wish to establish in vitro systems in which symmetry breaking in the 2D MT network can be reconstituted.

1SIP-01 アクチン系細胞骨格の SIM 及び STED による観察

Observation of actin-based cytoskeletal structures with SIM and STED microscopy

Kaoru Katoh[1], Keijyu Kamijo[2] ([1]*Biomed. Res. Inst., AIST*, [2]*Dept. Anat. & Anthropol., Tohoku Univ. Sch. of Med.*)

Super-resolution microscopes reveal fine structures smaller than resolution limits of conventional optical microscopy. We evaluated potential of SIM and STED microscope by observing actin meshwork of neuronal growth cones. STED was suitable for observation of fine structures (40-60nm) of fixed samples. In 3D stacks of STED, upper side and lower side of the thin growth cone were recognized as different optical slices (140nm thick in each slice). SIM was suitable for live cell imaging. Moreover, we determined fixation and staining condition suitable for SIM and STED.
We, therefore, used super-resolution to observe contractile ring of dividing cells. Arrangement of actin and myosin in the contractile ring will be shown in the presentation.

1SIP-02 構造化照明顕微鏡ＳＩＭの原理

Principle of the structured illumination microscopy

Yuki Terui (*Nikon*)

Recently, various methods of super resolution optical microscopy, which can break a barrier of classical diffraction limit, have been widely investigated. Some of these methods have already been commercialized and been used as efficient tools for many biologists who aim to clarify subcellular structures and their functions. Structured illumination microscopy (SIM) is one of these methods. By illuminating fringe pattern and analyzing multiple images, resolution is improved twice as high as conventional microscopy. Among several methods of super resolution microscopy, SIM has significant advantages in low photo toxicity, fast imaging speed, and a number of usable fluorophores. In this presentation, I will review the principle of SIM.

1SIP-03 リアルタイム超解像イメージングに向けた高速多重蛍光分子アルゴリズム Wedged Template Matching（WTM）の開発
Development of super resolution multi-emitter algorithm using template matching; towards real time analysis and visualization

Tomochika Takeshima[1], Teruo Takahashi[1], Jiro Yamashita[1], Yasushi Okada[2], **Shigeo Watanabe**[1] ([1]*Hamamatsu Photonics K.K.*, [2]*RIKEN QBiC*)

To improve the temporal resolution of single molecule switching nanoscopy, multi-emitter fitting algorithms have been developed. However they are computationally intensive.
Here we developed a rapid computational method, Wedged Template Matching (WTM), an algorithm using a template matching technique to localize highly overlapping fluorescent molecules with sub-diffraction resolution. WTM achieves localization of overlapping molecules at up to 20 molecules/μm^2 density with localization precision of 35 nm (RMSD) with both high detection sensitivity and fast computational speed.
WTM can resolve live cell protein dynamics with temporal resolution of several hundred milliseconds. The WTM computational speed enables real time-super resolution imaging during image acquisition.

1SIP-04 ピンホールを超えた共焦点超解像顕微鏡システム Airyscan
Airyscan: a new confocal based superresolution microscopy

Toshiyuki Watanabe[1], Klaus Weisshart[2] ([1]*Carl Zeiss Microscopy*, [2]*Carl Zeiss Microscopy GmbH*)

A lot of superresolution microscopies have been developed and several systems of them have been released from manufacturers as a product nowadays. However it's difficult to develop the microscopy which achieves both the superresolution and the enough versatility being able to handle a lot of applications simultaneously. To solve the issue, Carl Zeiss Microscopy introduced Airyscan, a new detector concept for laser scanning microscopy, which has several unique features and good versatility compare with other existing superresolution microscopies.
In this presentation, we introduce the application data taken by Airyscan and talk about the prospects in the near future.

1SIP-05 超解像蛍光顕微鏡法が明らかにするストレス顆粒内内在性 mRNA のナノスケール空間構成
Super-resolution fluorescence microscopy reveals nanoscale spatial organization of endogenous mRNA in stress granules

Ko Sugawara (*Grad. Sch. Pharm. Sci., Univ. of Tokyo*)

Under environmental stress, eukaryotic cytoplasmic mRNAs assemble to form stress granules (SGs). It is known that these densely-packed micrometer-sized granules regulate mRNA metabolism. However, the underlying mechanisms of this regulation remain unclear due to insufficient spatial resolution of conventional optical microscopy. Here, we investigated the spatial organization of endogenous mRNAs in SGs by localization-based super-resolution fluorescence microscopy, which achieved ~20 nm resolutions. Super-resolution imaging revealed that endogenous mRNAs form high-density clusters with a diameter of ~100 nm in single SGs. Our results suggested that SGs have highly organized structures that could be significant for the physiological functions in cellular stress responses.

1SIP-06 超解像イメージングで解き明かす植物細胞膜切断装置のダイナミクス
Directional assembly of plant endocytic dynamin unveiled by super-resolution imaging

Masaru Fujimoto[1], Akihiko Nakano[2,3], Takashi Ueda[2,4] ([1]*Grad. Sch. Agric. Life Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*RIKEN Center for Advanced Photonics*, [4]*PRESTO, JST*)

Dynamin has been proposed to assemble into ring-shaped structures at the neck of endocytic buds from the plasma membrane (PM), and to constrict and sever the neck; however, this has never been observed *in vivo*. In this study, we applied variable incidence angle fluorescence-combined structured illumination microscopy and super-resolution confocal live imaging microscopy with super spatial resolution, to observe the behavior of fluorescently-tagged plant dynamins. We demonstrated that plant dynamins assemble into ring-shaped structures preferentially in a clockwise direction, gradually concentrated into smaller foci, and finally separating from the PM. Our findings provide the first indication that dynamin directionally polymerizes into ring-shaped structures *in vivo*.

1SIP-07 Imaging local sphingomyelin domains in the plasma membrane using lipid-specific probes and super-resolution microscopy

Toshihide Kobayashi, Mitsuhiro Abe (*RIKEN*)

Lipid rafts are heterogeneous membrane domains composed of sphingomyelin (SM), glycosphingolipids and cholesterol, together forming the rigid structures in the plasma membrane. The proposed size of the raft is in the order of 10-200 nm, which is under the diffraction limit. Several SM-binding proteins have been characterized as probes to visualize SM in cell membranes. Combination of super-resolution optical microscopy and the SM-binding proteins enabled to visualize SM-enriched domains in living cells in sub-micron scale. Our results revealed the heterogeneity of SM domains on the cell surface and the role of SM-rich lipid rafts on regulating intracellular lipid distribution.

1SIP-08 Meiotic chromosome structure and function visualized with super-resolution microscopy

Peter Carlton (*iCeMS, Kyoto Univ.*)

In meiosis, homologous chromosomes must align along their lengths, and become connected by the synaptonemal complex (SC), a protein polymer. The width of the SC is less than 200nm, which limits its observation by fluorescence microscopy. We have used structured illumination microscopy to observe the SC at less than 200nm resolution, and characterized the SC in meiotic mutants that fail to complete meiotic pairing and recombination. Additionally, we show that the helical pitch of the SC is higher at the ends than in the middle of chromosomes, and increases over time. The SC structure is ideally suited to the size scale made accessible by superresolution microscopy, and future improvements will increase our understanding of this important macromolecular structure.

2SAA-01 タンパク質結晶内に創りだした隙間を利用して、タンパク質分子内部の運動分布を解析する

Crystal contact-free space for analyzing spatial distribution of protein internal motions

Daisuke Kohda (*Struct. Biol., Med. Inst. Bioreg., Kyushu Univ.*)

The strong non-linear distance dependency of data obtained by existing experimental techniques hinders the proper analysis of internal motions of protein molecules when large amplitude motions exist. In contrast, since the electron density is superimposable, an unbiased spatial view of motions is expected by crystallography, but only if the mobile parts are fortuitously located at the site lacking direct contacts with neighboring molecules in the crystal. Here we propose a strategy for designing a fusion protein to create crystal contact-free space (CCFS) in protein crystals and place the fluctuating segment/ligand in the created CCFS. We applied the CCFS method to visualize a ligand moving in the binding site of a protein, and a flexible segment in anther protein.

2SAA-02 蛋白質の分子揺らぎと構造変化を計測する X 線 1 分子動態計測法の開発

Refinements of the Diffracted X-ray Tracking Method for Recording Molecular Fluctuations and Conformational Changes of Proteins

Hirofumi Shimizu (*Univ. Fukui. Fac. Med. Sci.*)

We have been studying the structural dynamics of the KcsA potassium channel by the Diffracted X-ray Tracking (DXT) method in which the motions of a target protein were recorded as those of the diffraction spots from a gold nanocrystal securely attached to the protein. We have introduced a laser-triggered recording system, an x-ray focusing mirror, and a high-speed camera system, which enabled to record the gating motions and the enhancement of the molecular fluctuations prior to the opening conformational changes in a sub-millisecond time resolutions. In this session we will present recent progress of the method and discuss about its advantages.

2SAA-03 細胞内クラウディング環境における蛋白質のフォールディングとダイナミクスを NMR で観察する

NMR approaches to investigate protein folding and dynamics in the crowded intracellular environment

Yutaka Ito[1,2] ([1]*Tokyo Metropolitan Univ.*, [2]*CREST/JST*)

The in-cell NMR studies by Inomata et al. suggested that protein folding of ubiquitin in HeLa cells is destabilised, partly due to interactions with endogenous interacting proteins. This result was a challenge to the general belief that protein folding inside cells is stabilised through macromolecular crowding and macromolecular confinement effects. In the presentation, findings on the relationship between protein structure, dynamics and stability in intracellular environments will be briefly overviewed. In addition, I will present our recent in-cell NMR studies on the backbone dynamics of proteins inside E. coli cells and the effect of macromolecular crowding on the fold/unfold equilibrium of the drosophila drk N-terminal SH3 domain.

2SAA-04 Cytoplasmic conformational transition of Sec translocon

Yoshiki Tanaka[1], Yasunori Sugano[1], Mizuki Takemoto[2], Takaharu Mori[3], Takamitsu Haruyama[4], Arata Furukawa[1], Tsukasa Kusakizako[2], Kaoru Kumazaki[2], Ayako Kashima[2], Ryuichiro Ishitani[2], Hiroki Konno[4], Yuji Sugita[3], Osamu Nureki[2], **Tomoya Tsukazaki**[1,5] ([1]*NAIST*, [2]*Grad. Sch. of Sci., Univ. of Tokyo*, [3]*RIKEN*, [4]*BioAFM-FRC, Kanazawa Univ.*, [5]*JST, PRESTO*)

Protein translocation is one of the evolutionally conserved, fundamental mechanisms in all cells. E. coli SecYEG complex forms a protein-conducting channel. Membrane associated cytosolic protein SecA ATPase repeatedly pushes preprotein into the SecYEG channel in synchronization with ATP hydrolysis and then completes the protein translocation. Based on the crystal structures of each Sec component, functional studies of Sec machinery have been developed. However, dynamic interactions between Sec factors and its conformational changes still remain unclear. Here we report new crystal structures of SecYEG, suggesting conformational transition of the cytoplasmic region. We would also like to introduce our recent trials for visualizing the protein translocation process.

2SAA-05 大規模構造変化を伴う反応モデル構築のためのマルチレゾリューションシミュレーション手法の開発

Development of multi-resolution simulation methods for reactions with large conformational changes in biological system

Chigusa Kobayashi[1], Yasuhiro Matsunaga[1], Jaewoon Jung[1,2], Yuji Sugita[1,2,3,4] ([1]*RIKEN, AICS*, [2]*RIKEN, TMS*, [3]*RIKEN, iTHES*, [4]*RIKEN, QBiC*)

Recently, molecular dynamics (MD) simulations of biomolecules have been applied to elucidate the relationship between dynamics and biological functions. We have recently developed new high-performance MD software, GENESIS, for the efficient simulations on K computer and PC clusters. Large conformational changes in response to reactions in biological systems, however, are still difficult to be simulated using all atom force fields due to the long time-scale of motions. To overcome the difficulty, we have developed new structure-based coarse-grained (CG) models describing conformational change between different states. We discuss novel conformational sampling algorithms by using the CG models for biomolecular simulations.

2SAA-06 The identification of the ankyrin repeat domain as a novel lipid-binding module

Shiro Suetsugu[1], Nobuaki Takahashi[2], Yuzuru Itoh[3], Kazuhiro Takemura[3], Akio Kitao[3], Yasuo Mori[2] ([1]*Graduate School of Biological Sciences, Nara Institute of Science and Technology*, [2]*Graduate School of Engineering, Kyoto University*, [3]*Institute of Molecular and Cellular Biosciences, University of Tokyo*)

The diversity of protein modules for lipid binding is not well established. Here we screened the lipid binding domains and found that the ankyrin-repeat domain (ARD) can bind to lipid. In particular, we focused on the ARD of TRPV4, because mutations in the ARD of TRPV4 are responsible for several channelopathies. Among the phosphoinositides, phosphatidylinositol-4,5-bisphosphate (PI(4,5)P2) most potently binds to the TRPV4 ARD. The crystal structure of the TRPV4 ARD in complex with inositol-1,4,5-trisphosphate, the head-group of PI(4,5)P2, and the molecular-dynamics simulations revealed the PI(4,5)P2-binding amino-acid residues. Notably, disease-associated TRPV4 mutations that cause a gain-of-function phenotype abolished PI(4,5)P2 binding and PI(4,5)P2 sensitivity.

2SDA-01 膜蛋白質ー膜脂質の相互作用解析を目指した重元素化脂質の利用

Application of synthetic lipids for analysis of interaction between membrane protein and lipids

Shigeru Sugiyama[1,2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*JST, ERATO*)

The lipids binding to membrane protein play a crucial role in membrane protein organization and function. Therefore, a crystallographic study of the membrane protein-lipid complex is very important to elucidate the detailed mechanism of its lipid recognition. However, the conformation of lipids interacted with membrane protein is hardly determined. Membrane protein is crystallized upon solubilization by detergents. This treatment often disrupts innate structures and functions. Here we focused on the bicelle crystallization method. In this study, bacteriorhodopsin was crystallized using the bicelle formulations complexed with bromine-containing compounds. We propose the bicelle crystallization technique to gain deeper insight into biomembranes.

2SDA-02 脂質ラフトの NMR 解析

NMR Studies of Lipid Rafts

Nobuaki Matsumori[1], Michio Murata[2,3] ([1]*Grad. Sch. Sci., Kyushu Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*ERATO*)

Lipid rafts are membrane domains rich in sphingolipids and cholesterol (Chol), and are assumed to play essential roles in biological processes such as signal transduction. However, molecular basis of raft formation have yet to be elucidated. In this study, we took three NMR approaches to reveal the structure and dynamics of lipid molecules in rafts; the first one is conformation analysis of sphingomyelin (SM) using bicelles, the second is determination of orientation of SM amide plane using dipolar couplings and chemical shift anisotropies, and the third is dynamics analysis of SM alkyl chains using 2H NMR. Based on these data, we will discuss why SM preferentially forms lipid rafts phase rather than saturated glycerophospholipid in biomembranes.

2SDA-03 FABP3 脂肪酸結合における内部水分子の重要性

Interplay between Internal Water Molecules and Bound Fatty Acid Molecule in FABP3 Binding Cavity

Daisuke Matsuoka[1,2], Shigeru Matsuoka[1,2], Shigeru Sugiyama[1,2], Michio Murata[1,2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*ERATO Murata Project*)

Fatty acid binding protein 3 (FABP3) is involved in fatty acid (FA) cytosolic transport. Its internal binding cavity accommodates one FA molecule and also about 13 ordered water molecules. Recently, we demonstrated that the internal water molecules play a key role for the wide ligand selectivity of FABP3 through the characterization of the hydration structures in the binding cavity.
In this presentation, we examine the importance of the hydration structure for the stable ligand binding by FABP3. To identify scaffold residues for the H-bond networks, FABP3 mutants were prepared and their FA affinities were evaluated by ITC. Computational study indicated that these residues affect the binding stability via the hydration structure by either direct or indirect mechanism.

2SDA-04 Structural basis for glycolipid recognition of human immune receptor, Mincle, and rational inhibitor design

Atsushi Furukawa[1], Takanori Matsumaru[2], Risa Ikeno[1], Yusuke Shuchi[1], Sho Yamasaki[3], Toyoyuki Ose[1], **Katsumi Maenaka**[1,2] ([1]*Laboratory of Biomolecular Science, Faculty of Pharmaceutical Sciences, Hokkaido University*, [2]*Center for Research and Education on Drug Discovery, Faculty of Pharmaceutical Sciences, Hokkaido University*, [3]*Division of Molecular Immunology, Research Center for Infectious Diseases, Medical Institute of Bioregulation, Kyushu University*)

Human immune C-type lectin receptor, Mincle (Macrophage inducible C-type lectin, CLEC4E), recognizes cord factor, TDM (Trehalose-6,6'-dimycolate), a glycolipid of Mycobacterium tuberculosis cell wall, to induce adjuvant activity. Mincle is supposed to simultaneously interact both sugar and lipid moieties of TDM, based on our crystal structure and mutagenesis study of Mincle. We here report the analysis for binding of Mincle with modified lipid compounds. We synthesize the derivatives of several lipids harboring different set of acyl groups. The binding activity revealed the C12 acyl group is a minimum essential unit for Mincle recognition. We further perform the structural study for these interactions. These results will provide clues to rational design for effective Mincle-mediated adjuvants.

2SDA-05 新規部分フッ素化リン脂質膜の創製

Development of Novel Partially Fluorinated Phospholipid Membrane for Biophysical Studies of Membrane Proteins

Masashi Sonoyama (*Div. Mol. Sci., Fac. Sci. & Tech., Gunma Univ.*)

Phospholipid membrane is an essential platform for functional expressions of membrane proteins (MPs). Novel artificial phospholipid membrane that provides a structural framework for MPs in the functional state has been desired long for further progress in biophysical studies of MPs in vitro. Recently we have demonstrated that a novel partially fluorinated phosphatidylcholine (PC), F4-DMPC, an analog molecule of Dimyristoylphosphatidylcholine (DMPC) with the perfluorobutyl group in the hydrophobic tail, is a very promising material for stably incorporating MPs in the functional state. In this presentation, structural and functional properties on proteoliposome of bacteriorhodopsin and partially fluorinated PCs with different perfuluroalkyl chain lengths will be reported.

2SDA-06 膜孔形成毒素の自己組織化に生体膜の組成とダイナミクスが与える影響の分子基盤

Molecular basis of self-assembly of a transmembrane hemolytic toxin triggered by specific membrane composition and dynamics

Koji Tanaka[1], Jose Caaveiro[1], **Kouhei Tsumoto**[1,2] ([1]*School of Engineering, The University of Tokyo*, [2]*The Institute of Medical Science, The University of Tokyo*)

Pore-forming toxins (PFTs) are key molecular components of the defense and attack frontline system in living organisms. PFTs are water-soluble proteins that transform into transmembrane pores on the target lipid membrane, resulting in cell-damage. The molecular mechanism of membrane-specific attack of PFTs is, however, unclear. Herein we have elucidated the activation mechanism of the hemolytic protein fragaceatoxin C (FraC), a PFT by determining crystal structures of the transmembrane pore at 3.1 Å. The pore of FraC is composite particle composed of protein chains and sphingomyelin that act as assembly co-factors. In addition, it was revealed that domain formation of membranes kinetically facilitates pore-insertion to membranes.

2SFA-01 動物胚が発生と進化の法則性、砂時計モデルに従うのはなぜか？

What makes animal embryos to follow the hourglass model?

Naoki Irie (*Grad. Sch. Sci., Univ. Tokyo*)

Based on gene expression profiles during animal embryogenesis, recent studies (including our study) supported that developmental hourglass model best explains the relationship between animal embryogenesis and evolution. Counterintuitively, hourglass model predicts that earliest developmental stages are not conserved the most, but mid embryonic stages, when variety of cells exist, are the period of strongest conservation.
But why? Scientists are now actively discussing the possible constraints by "developmental system" itself. Why embryonic stages with variety of cells should be conserved than single-celled, fertilized eggs?
Main focus of this talk would be the discussion on possible evolutionary mechanism that makes embryos to follow the hourglass model.

2SFA-02 腸内細菌叢由来代謝産物がもたらす生体恒常性維持機構

Gut microbiota-derived metabolites shape host physiological homeostasis

Shinji Fukuda (*Inst. Adv. Biosci., Keio Univ.*)

The gut microbiota form a highly complex ecological community together with host intestinal cells. The so-called gut ecosystem has a profound influence on human physiology. It has been reported that imbalance in the gut ecosystem could be a risk factor in human disorders including not only gut-associated disorders, but also systemic diseases. However, the molecular basis of the gut microbiota function through host-microbial crosstalk remain obscure. To this end, we established a novel omics-based approach which integrates metagenomics and metabolomics, and found that gut microbiota-derived metabolites largely affected to the host phenotype, suggesting that gut microbiota-derived metabolites are considered to be therapeutic target molecules for human disorders.

2SFA-03 シングルセル遺伝子発現解析が明らかにする細胞分化のメカニズム

Single Cell Transcriptome Analysis Dissects Cell Fate Specification

Akira Watanabe (*Kyoto University*)

A human single-cell fertilized egg generates hundreds of different types of the cell by epigenetic mechanism. To address the mechanisms by which a particular gene network at a branch of the cell differentiation defines direction to specific cell type, we address detailed dynamics of transcription during differentiation from induced pluripotent stem (iPS) cells to cardiomyocyte. We performed single cell RNA-seq using iPS and cardiomyocyte-directed cells. Our newly developed bioinformatical approach identified dynamics ofntranscriptome during the differentiation process. We propose time-information-free analysis as a powerful approach for unveiling the dynamics of transcriptome in reprograming and differentiation.

2SFA-04 Dynamics of phenotype-genotype mapping in laboratory evolution of Escherichia coli

Chikara Furusawa (*QBiC, RIKEN*)

Laboratory evolution is a powerful tool to quantify dynamics of phenotype-genotype mapping, which enables us to unveil the nature of adaptive evolution. In this study, we performed laboratory evolution experiments of E. coli under various conditions, and expression changes and genomic changes were quantified by microarray analysis and next-generation sequencing, respectively. The results of these comprehensive analyses demonstrated that the expression changes were restricted to a low-dimensional dynamics, while diverse genomic changes can contribute to similar phenotypic changes. A non-genetic memory contributing adaptive phenotypic changes was also suggested. Based on these results, universal features in adaptive evolution will be discussed.

2SFA-05 進化と適応における揺らぎ、応答の普遍関係

Fluctuation and Response in Adaptation and Evolution-Universal Relationship

Kunihiko Kaneko (*Univ. of Tokyo, Departmet of Basic Science*)

Characterization of plasticity, robustness, and evolvability is an important issue in biology. First, we present a macroscopic theory of fluctuation and responses in expressions across genes, by assuming that cells undergo steady growth. From the constraint that all components have to be approcimately doubled for reproduction, expression of all genes is shown to change along a one-parameter curve in the state space, in response to the environmental stress. Second, proportionality between phenotypic variances due to environmental noise and to genetic variation is formulated by macroscopic phenomenology theory, as is supported by numerical evolution.
Last, we discuss how consistency between evolutionary and developmental scales leads to these universal laws.

2SGA-01 高速 AFM で明らかにする分子シャペロン ClpB ーの柔らかさと機能

Structural Flexibility and Chaperone Activity of ClpB observed by High-Speed AFM

Takayuki Uchihashi[1,2,3], Yo-hei Watanabe[4], Ryota Iino[5], Toshio Ando[1,2,3] ([1]*Coll. Sci. & Eng. Kanazawa Univ.*, [2]*Bio-AFM FRC, Kanazawa Univ.*, [3]*CREST-JST*, [4]*Dep. Biol., Konan Univ.*, [5]*OIIB/IMS*)

A molecular chaperon ClpB is a ring-forming ATPase that rescues aggregated proteins. Conformational changes of ClpB in a hexameric ring are considered to be indispensable for disaggregation activity but the detailed mechanism remains unknown. We applied HS-AFM to directly visualize conformational dynamics of the ClpB ring. The HS-AFM observation revealed that the ClpB ring is highly flexible: the ring repeats reversible opening and closing and even number of the subunits which consists the ring fluctuates in an ATP dependent manner. Furthermore, most of the ring was open for the hyper-active mutant while the flexibility was repressed for the less-active mutant. Our results indicate dynamic flexibility of the ClpB ring plays essential roles for protein disaggregation.

2SGA-02 Theoretical study of ion transport pathway of channelrhodopsin

Norio Yoshida (*Kyushu Univ.*)

Channel-rhodopsin (ChR) is known as a light-gated ion channel, which is derived from unicellular green algae. Recently, the crystal structure of a ChR in the closed dark-state has been revealed by Nureki and co-workers, they also suggested the cation conduction pathway. However, the open-state structure and its cation conduction pathway are still unknown. In the present study, we applied the statistical mechanics theory of molecular liquids called three-dimensional reference interaction site model (3D-RISM) metheod and molecular dynamics simulation to investigate the cation conduction pathway of ChR in both closed/opened-state.

2SGA-03 固体 NMR によるファラオニスフォボロドプシンの Tyr174 と Tyr199 の水素結合変化の解析

Solid-state NMR study of hydrogen-bonding alterations of Tyr174 and Tyr199 in pharaonis phoborhodopsin

Izuru Kawamura (*Grad. Sch. Eng., Yokohama Natl. Univ.*)

Pharaonis phoborhodopsin (*p*pR) is a seven-helical transmembrane photoreceptor with a retinal chromophore and forms a 2:2 complex with its cognate transducer protein, *p*HtrII, in the membrane. A hydrogen bond between Tyr174 and Thr204 in the retinal-binding pocket plays an essential role to express a negative phototaxis function. Tyr199 in the transmembrane region of *p*pR also plays an important role to interact with *p*HtrII. Here, we have demonstrated sharp alterations of hydrogen bonds of Tyr174 and Tyr199 in site-specific ^{13}C isotope labeled *p*pR reconstituted into fully hydrated EggPC lipid bilayers by ^{13}C solid-state NMR at 300 K. We will discuss about the alterations of hydrogen bonds in such soft molecular system.

2SGA-04 カリウムイオンを感知して活性がスイッチングするインテリジェントリボザイム/アプタマーの創製

Development of intelligent ribozyme/aptamer that sense K^+ and switch on their activities

Masato Katahira (*Inst. Adv. Energ., Kyoto Univ.*)

The RNA strand, r(GGAGGAGGAGG) (R11), takes on an extended single-strand in the absence of K^+, while it folds into a compact quadruplex in the presence of K^+. We divided the active core of a ribozyme into two subunits, and they were joined to each end of R11, respectively. The constructed molecule is inactive in the absence of K^+, while in the presence of K^+, R11 forms the compact quadruplex and thus brings two subunits close to each other, resulting in the restoration of the active core structure, and thus this molecule exerts enzymatic activity. In this way, this molecule can switch on its enzymatic activity through sensing of K^+. With the same strategy, we have also developed an intelligent aptamer that switches on its protein-trapping activity in response to K^+.

2SGA-05 蛋白質分子の機能的運動を制御するナノスケールの水和構造変化

Nanoscale wetting and drying processes dominate protein functional motions

Tomotaka Oroguchi, Masayoshi Nakasako (*Sci. Tech., Keio Univ.*)

Water is ‘mother liquid’ for life. For example, aqueous environment induces protein structural flexibilities indispensable for its function. Here we visualized that the protein functional motions are dominated by dissociation and association of a small number or hydration water molecules in hydrophobic and hydrophilic pockets. Using molecular dynamics simulation and experiments, we identified the functional domain motion of an enzyme to open/close its active-site cleft in solution. The atomic-detailed analyses on the conformational and hydration changes reveal that ‘wetting’ and ‘drying’ processes occurring at the cleft control the domain motion. Our results demonstrate how water molecules play crucial roles in large-scale and functional protein motions.

2SGA-06 立体選択的な水酸化酵素をめざしたマンガンポルフィセン錯体を含むミオグロビンの設計と構築

Design and engineering of myoglobin containing a manganese porphycene toward enantioselective hydroxylase

Koji Ohora (*Grad. Sch. Eng. Osaka Univ.*)

Myoglobin (Mb), an oxygen storage hemoprotein containing heme b, does not catalyze alkane hydroxylation. In contrast, cytochrome P450 including the same cofactor shows the high hydroxylase activity. In this decade, our group has demonstrated artificial metalloenzymes by hemoprotein reconstitution with artificial cofactors. Recently, we have focused on Mb reconstituted with a Mn porphycene complex (MnPc) to catalyze alkane hydroxylation due to C-H bond activation property of a Mn complex. Mb reconstituted with MnPc catalyzes the H_2O_2-dependent hydroxylation of ethylbenzene. In this presentation, we will report the molecular dynamics simulation-based design of Mb mutants for the reconstituted protein toward high enantioselectivity.

2SHA-01 Single-molecule analysis of conformational space of a yeast prion protein Sup35NM using optical tweezers

Yusuke Komi[1], Rodrigo Maillard[2], Piere Rodriguez[2], Carlos Bustamante[2,3], Motomasa Tanaka[1] ([1]*BSI, RIKEN*, [2]*UC Berkeley*, [3]*HHMI*)

Protein conformation fluctuates dynamically with different aspects of conformation changing. The conformational diversity of monomer has a strong impact on the process of protein folding and misfolding. Here we investigate the conformation space of an intrinsically disordered protein Sup35NM by single-molecule force measurement using optical tweezers. We found that the conformational space of the N-terminal prion domain in Sup35NM was different from that of full-length Sup35NM. This result suggest that there is the difference in aggregation propensity between a nascent polypeptide and a mature form of Sup35NM. Furthermore, our structural analysis for a Sup35NM mutant revealed how conformational space provides an effect on formation of a specific amyloid conformation.

2SHA-02 タンパク質翻訳の１分子研究
Single molecule study of protein translation

Sotaro Uemura (*Graduate School of Science, The University of Tokyo*)

Translation by the ribosome occurs by a complex mechanism involving the coordinated interaction of multiple nucleic acid and protein ligands. Here we use zero-mode waveguides (ZMWs) and sophisticated detection instrumentation to allow real-time observation of translation at physiologically ligand concentrations. Translation at each codon is monitored by stable binding of tRNAs—labelled with distinct fluorophores—to translating ribosomes. We observe the transit of tRNAs on single translating ribosomes and determine the number of tRNA molecules simultaneously bound to the ribosome. Our results show that ribosomes are only briefly occupied by two tRNA molecules and that release of deacylated tRNA from the exit (E) site occurs rapidly after translocation.

2SHA-03 SecM による翻訳停止の分子メカニズムに関する研究
Study on the molecular mechanism of translation arrest by SecM

Zhuohao Yang, Ryo Iizuka, Yuanfang Guo, **Takashi Funatsu** (*Grad. Sch. Phar. Sci., Univ. Tokyo*)

SecM, a bacterial secretary protein, contains at its C-terminus an arrest sequence which interacts with the ribosomal tunnel and arrests its own translation. We performed in vitro translation assays with HaloTag proteins fused to the C-terminal fragment containing the arrest sequence or the full-length SecM, and found that the nascent SecM outside the ribosome markedly stabilizes the translation arrest. These results indicate that changes in the nascent polypeptide chain outside the ribosome can affect the stability of translation arrest. Next we examined whether translation could be rescued by pulling SecM-ribosome complex by optical tweezers, because release of arrest at the translation has been suggested to occur by mechanical force exerted by the translocon.

2SHA-04 Incorporation of fluorescent amino acid for lifetime FRET

Takanori Uzawa (*RIKEN*)

Successful protein translation is the essential process in living organisms. Although the powerful translation system in experimental cells is routinely utilized to obtain desired proteins, our knowledge on the very first step of the translation is still limited. For example the knowledge on the nascent-chain folding on ribosome has not been accumulated as much as that on the protein refolding from denaturing conditions. Thus motivated we are constructing a lifetime FRET system; we first incorporate a FRET pair of non-natural, fluorescent amino acids at specific positions of a nascent chain on ribosome and monitor the change in donor lifetime in presence and absence of acceptor. We plan to apply this system for the nascent-chains of various proteins.

2SHA-05 シャペロニン空洞内フォールディングにおける疎水性相互作用の役割
The role of hydrophobic tethering in chaperonin-mediated folding

Fumihiro Motojima[1], Masasuke Yoshida[2] ([1]*Biotech. Res. Cent. and Dept. of Biotech., Toyama Pref. Univ.*, [2]*Kyoto Sangyo Univ.*)

Chaperonins fold protein in the central cavity formed by the large oligomeric ring complex. However, the mechanism by which a chaperonin mediates protein folding is still controversial. Recently, we found that denatured proteins associate with hydrophobic residues in the chaperonin cage (tethering), and those in the middle of the cage are especially important for both retaining and folding denatured protein in the cage. These results suggest that hydrophobic tethering is necessary to fold denatured proteins isolated in the cage.

2SHA-06 高速 AFM が明らかにする PDI の酸化的フォールディングの触媒機構の解明
A new mechanism of operation of PDI during its catalysis of oxidative protein folding revealed by high-speed atomic force microscopy

Masaki Okumura[1], Kentaro Noi[2,5], Shingo Kanemura[1], Shoji Masui[1], Takaaki Hikima[3], Shuji Akiyama[3,4], Teru Ogura[2,5], Kenji Inaba[1,5] ([1]*IMRAM, Tohoku University*, [2]*IMEG, Kumamoto University*, [3]*RIKEN Harima*, [4]*IMS*, [5]*CREST, JST*)

PDI is an efficient catalyst that introduces native disulfide bonds into secretory proteins in the endoplasmic reticulum. In this study, we visualized actions of PDI during its catalysis of disulfide bond formation at the single-molecule level by employing high-speed atomic force microscopy (HS-AFM). The HS-AFM analysis revealed that PDI undergoes significant redox-dependent regulations of the thioredoxin domain arrangement; oxidized form of PDI is in dynamic equilibrium between “open” and “closed” conformations whereas the reduced form holds a “closed conformation” persistently. Real-time monitoring of PDI when capturing an unfolded substrate is now in progress.

2SHA-07 小胞体トランスロコンによる新生鎖のハンドリング
Handling of nascent chain by ER translocon during membrane protein integration

Masao Sakaguchi (*Grad. Sch. Life Sci., Univ. Hyogo*)

Membrane proteins are cotranslationally integrated into the endoplasmic reticulum membrane via the protein-conducting channel, the so-called translocon. During the process, amino acid sequences of nascent chains elongating from ribosome are scanned by the channels and classified to be retained in the membrane or translocated into the lumen. The classification processes are fundamental one for the membrane protein folding. Positive charges on the nascent chain slowdown the movement of nascent chain though translocon. Even marginally hydrophobic segments are maintained as transmembrane segment in the translocon and the handling is regulated by the ribosome. Interestingly, nascent chain within the ribosome tunnel greatly affects the handling function of translocon.

2SIA-01 ３つのタンパク質で再構成できるシアノバクテリアの生物時計

A self-sustaining and temperature-compensated circadian rhythm reconstituted *in vitro* in a minimal system containing three Kai proteins

Kazuki Terauchi (*Dep. Life Sci., Ritsumeikan Univ.*)

The cyanobacterial circadian oscillator can be reconstituted by mixing purified clock proteins, KaiA, KaiB and KaiC with ATP to exhibit a 24-h oscillation of phosphorylation of KaiC *in vitro*. The reconstituted circadian clock satisfies the criteria of circadian rhythms: persistence in constant conditions, phase resetting and temperature compensation of the period. KaiC is a hexameric protein exhibiting ATPase, autophosphorylation and autodephosphorylation activities. KaiA stimulates the autophosphorylation of KaiC, whereas KaiB inhibits the effect of KaiA. In this symposium, I will present recent experiments addressing the molecular mechanism underlying the cyanobacterial circadian clock.

2SIA-02 KaiC as Circadian Pacemaker of Cyanobacterial Circadian Clock

Shuji Akiyama (*Institute for Molecular Science*)

KaiC is a core protein of the cyanobacterial Kai oscillator, which persists without transcription-translation feedback. In the presence of KaiA and KaiB, KaiC reveals rhythmic activation/inactivation of its ATPase and autokinase/autophosphotase activities over approximately 24 h. Newer data are emerging on its slow, temperature-compensated ATPase activity, which is involved in the pacemaker for dynamic cellular events such as assembly/disassembly of the Kai components in cyanobacteria. To seek the molecular basis of the circadian timescale, we have studied the structure and function of KaiC ATPase extensively. On the basis of recent observations, we will discuss the origins of slow but ordered dynamics of the Kai oscillator.

2SIA-03 KaiABC システムにおける ATP 加水分解と概日振動の結合についてのメゾスケールモデリング

Mesoscale modelling of coupling between ATP hydrolysis and circadian oscillation in KaiABC system

Sumita Das, Shota Hashimoto, Tomoki P. Terada, **Masaki Sasai** (*Department of Computational Science and Engineering, Nagoya University*)

KaiABC system is unique in its ability to show the stable circadian oscillation in vitro, but its oscillation mechanism is still elusive. This problem should be decomposed into three levels. In the macroscopic level with the size of test tube, many KaiC molecules must synchronize in their oscillation phase to show the observable oscillation, and in the microscopic level, atomic structural change is responsible for two types of reactions, ATP hydrolysis and phosphorylation/dephosphorylation. In the mesoscopic level, these two types of reactions should be coupled to each other in single KaiC hexamer giving feedback regulation of each other. We discuss the stochastic simulation modelling in this mesoscopic level to reveal the role of ATP hydrolysis in oscillation.

2SIA-04 ATP 駆動分子モーターの１分子高速可視化解析

Single-molecule high-speed imaging analysis of ATP-driven molecular motors

Ryota Iino (*OIIB and IMS, NINS*)

ATP-driven molecular motors convert chemical energy of ATP hydrolysis into unidirectional mechanical motion. This energy conversion generally accompanies changes in conformations of the subunits which constitute the motors. We are trying to elucidate conformational dynamics and transient intermediate structures of moving molecular motors with high-speed single-molecule imaging probed by gold nanoparticles and gold nanorods. The detail of motions such as mechanical steps generating force and pauses waiting for elementary steps of chemical reaction, and conformational changes inside the individual molecular motors can be monitored. In my talk, I will introduce our recent data on a linear molecular motor kinesin and rotary molecular motors F- and V-ATPases.

2SIA-05 細胞の力覚応答と増殖におけるアクチン動的秩序の役割

Roles of actin dynamic ordering in mechanosensing and cell proliferation

Kensaku Mizuno (*Grad. Sch. Life Sci., Tohoku Univ.*)

Actin turnover is one of the major ATP-consuming processes in cells. Actin dynamics play essential roles in cell morphology, migration, proliferation and mechanosensing. Mechanical forces applied to cells induce actin cytoskeletal reorganization, such as stress fiber reinforcement, allowing cells to protect from the forces. However, how cells respond to forces remains elusive. We identified a RhoA-GEF, Solo (ARHGEF40), that is involved in cyclic-stretch-induced endothelial cell reorientation. We also found that Solo interacts with keratin filaments and this interaction plays critical roles in tensile force-induced RhoA activation and stress fiber reinforcement. We also provide evidence that actin dynamics regulate cell proliferation and primary cilium formation.

2SIA-06 生体溶液中での ATP 加水分解による同符号電荷間引力制御と時空間動秩序

Spatiotemporal dynamic ordering regulated by ATP hydrolysis and effective attraction between negatively charged sites in a biofluid

Ryo Akiyama (*Dept. of Chem., Kyushu Univ.*)

We have studied strong effective attraction between macroanions immersed in electrolyte solution by using HNC-OZ theory, which is an integral theory. When the “macroanions” have oxygen sizes and electronic charges, some interesting behaviors are observed in the effective interaction between the “macroanions”, namely O-. The strong attraction appears if the mediators of attraction are multivalent cations and the electrolyte concentration is about 10-2 M. These calculated results agree with experimental results for a solution of acidic proteins, qualitatively. Moreover, the attraction becomes stronger as the charges of “macroanions” becomes larger. We will discuss spatiotemporal dynamic ordering regulated by ATP hydrolysis on the basis of those results.

2SJA-01 人工細胞の進化的動態
Evolutionary dynamics of an artificial cell model

Tetsuya Yomo[1,2] (*[1]Grad. Sch. Info. Sci. & Tech., Osaka Univ.*, *[2]Grad. Sch. Front. Bio-Sci., Osaka Univ.*)

The ability to evolve is a key characteristic that distinguishes living things from non-living compounds. We encapsulated an artificial RNA genome and the factors for protein synthesis into water droplets in oil or lipid vesicles. In the micro-compartments, the artificial genomic RNA produced a RNA replicase, which in turn replicates the original RNA. The micro-compartment population evolved as if it were natural cells. The population gained mutations through the error in the replication of RNA and took the advantage of natural selection. Beyond the simple optimization by natural selection, they showed host-parasite dynamics and hitchhike-type molecular evolution. Some similarity of the artificial cell model to natural cells will be discussed.

2SJA-02 二成分脂質ベシクルで探る生命と分子集合体の境界
A border between cellular life and molecular assembly revealed by binary lipid vesicles

Yuka Sakuma (*Grad. Sch. Sci., Univ. Tohoku*)

Evolution of molecular assemblies toward a cellular life is fascinating research filed in soft matter science. In order to bridge between the molecular assemblies and the cellular life we should create model assembly systems having fundamental functionalities for cellular life and reveal the underlying physics to reproduce these functionalities in the molecular assemblies. We have succeeded to reproduce the various vesicle deformations relevant to the cell functionalities, such as adhesion, pore formation, and self-reproduction, using simple binary vesicles. The heart of these shape deformation mechanisms is the interplay between vesicle area regulation via the chain melting of lipids and the localization of lipids with the preference spontaneous curvature.

2SJA-03 バクテリアサイボーグ
Bacterial cyborg; integrated bacterial cell systems into arrayed lipid bilayer chamber

Hiroyuki Noji (*School of Engineering, The University of Tokyo*)

We developed bacterial cyborg by the integration of E. coli protoplasts with arrayed lipid bilayer chambers (ALBiCs), by membrane fusion. The inner volume of ALBiCs is 10-30 fL and comparable to that of E. coli protoplast, 10-20 fL. Thereby, the cytosol of protoplast is diluted only a few times upon the fusion. The viability of bacterial cyborg was tested from gene expression activity and membrane morphological change. Although the gene expression activity of bacterial cyborg were remarkably weak compared with in vitro translation in femtoliter chambers, some chambers showed drastic membrane morphological changes such as extrusion and budding, suggesting some bacterial cyborg retained viability.

2SJA-04 ゲノム合成生物学でのゲノム構築
Scrapping and Building Bacteria Genomes for Novel Synthetic Genomics

Mitsuhiro Itaya (*Inst. Adv. Biosce. Keio Univ.*)

A eubacterium called *Bacillus subtilis* 168 has been able to construct giant DNA as large othor bacterial genomes, since the first successful report highlighted by the stable cloning of a whole 3.5-Mb genome of the nonpathogenic, unicellular photosynthetic bacterium *Synechocystis*. It is suggested that most bacteria genomes would be targets for cloning in this host regardless of existing or newly designed. Despite of real genome construction/cloning, we need breaks how to make genomes function in a new cellular bag called chassis in synthetic biology field. Our current scenarios and unprecedented methods being developed will be presented. They might lead to new pipelines that should provide functional genomes for novel synthetic biology fields.

2SJA-05 Mutation accumulation of bacterial genome toward genomic inactivation

Saburo Tsuru, Atsushi Shibai (*Osaka Univ.*)

Modern living cells consist of many kinds of molecular machines which have already evolved to obtain their sophisticated functions and shapes. To explore a primitive form of life without such evolved compositions, it is useful to degenerate existing organisms by accumulating destructive mutations on their genome. Using bacterial cells with high mutability and a mutagen, we have empirically tried to accumulate many mutations on the genome toward genomic inactivation. We report the mutation rate and mutational spectrum of the accumulation processes.

2SKA-01 Opn5L1 は光サイクル性の反応で制御される G タンパク質共役型受容体である
Opn5L1 is a photocyclic GPCR

Keita Sato[1], Takahiro Yamashita[1], Hideyo Ohuchi[2], Atsuko Takeuchi[3], Sayuri Tomonari[4], Kazumi Sakai[1], Yasushi Imamoto[1], Akimori Wada[3], Yoshinori Shichida[1] (*[1]Grad. Sch. Sci., Kyoto Univ.*, *[2]Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*, *[3]Kobe Pharm. Univ.*, *[4]Inst. Tech. Sci., Univ. Tokushima*)

Photoreceptive protein opsin is G protein-coupled receptor, which employs retinal as a chromophore ligand. Here we show that G protein activation efficiency of opsin named Opn5L1 is controlled by light-dependent suppression and subsequent thermal recovery. Opn5L1 is activated by binding of agonist all-trans-retinal, not by photoreception. All-trans-retinal-bound state of this opsin absorbs in the visible region. Upon light absorption, Opn5L1 is converted into a light-insensitive inactive state. This inactive state thermally reverts to the original all-trans-retinal-bound state within several hours, resulting in recovery of G protein activation efficiency. The detailed molecular mechanism of this cyclic reaction will be discussed.

2SKA-02 Evolution of the umami taste receptor in vertebrates

Yasuka Toda[1,2] ([1]*Kikkoman Corp.*, [2]*Dept. Appl. Biol. Chem., The Univ. Tokyo*)

Taste perception is essential for food selection. Among the five basic tastes, umami is sensed by a heteromeric complex of class C GPCRs termed T1R1 and T1R3. T1R1/T1R3 has been considered to be a sensor of protein sources, because the known natural ligands of this receptor are only L-amino acids. T1R1/T1R3 exhibits species-dependent differences in amino acid selectivity. We human beings specifically perceive L-Glu as having umami taste, although T1R1/T1R3 in the other species generally exhibits broadly tuned amino acid sensing properties. Here we show the molecular mechanisms underlying the L-Glu-specific responses in human T1R1/T1R3. Moreover, we describe a novel sweet taste detection mechanism that evolved in a nectar-feeding bird by changing function of T1R1/T1R3.

2SKA-03 膜型 TGFα 切断を利用した GPCR 活性化の簡便な検出法とその応用
TGFα shedding serves a useful readout for dissecting GPCR signaling

Asuka Inoue[1,2] ([1]*Grad. Sch. of Pharm. Sci., Tohoku Univ.*, [2]*PRESTO, JST*)

The GPCR family consists of more than 800 members in the human genome and serves as the most successful drug targets. Heterotrimeric G proteins (Gs, Gi, Gq and G12) mediate majority of GPCR actions. Thus, quantifying G protein coupling is important, yet difficult due to distinct downstream signaling. Here, we extended the assay to quantitatively measure G protein coupling of a GPCR. To this end, we generated G protein-deficient HEK293 cells using a CRISPR-Cas9 system. By measuring coupling and consequent downstream signaling of 11 chimeric Gα subunits by TGFα shedding assay (Inoue et al. Nat Methods 2012), we could determine a G protein coupling signature of its GPCR. The landscapes of G protein coupling will be useful for dissecting in vivo roles of GPCRs.

2SKA-04 NMR 法を用いた脂質二重膜中の GPCR の動的構造解析
Functional dynamics of G-protein-coupled receptors in lipid bilayers revealed by NMR

Yutaka Kofuku[1], Takumi Ueda[1,2], Junya Okude[1], Yutaro Shiraishi[1], Keita Kondo[1], Takuya Mizumura[1], Shiho Suzuki[1], Ichio Shimada[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*PRESTO, JST*)

G-protein-coupled receptors (GPCRs) exist in conformational equilibrium between active and inactive states, and the former population determines the efficacy of signaling. However, the conformational equilibrium of GPCRs in lipid bilayers is unknown. We utilized NMR to investigate the structure of β_2-adrenergic receptor (β_2AR) in lipid bilayers of reconstituted high-density lipoprotein (rHDL). Our results revealed that the exchange rates for the conformational equilibrium of β_2AR in rHDLs were remarkably different from those measured in detergents. The fast timescale of activation of intracellular signaling by GPCRs, as calculated from the exchange rates, enables rapid neurotransmission and sensory perception.

2SKA-05 バイオインフォマティクスによる GPCR オリゴマー研究
Bioinformatics approaches in the study of GPCR oligomers

Wataru Nemoto[1], Yoshihiro Yamanishi[2], Vachiranee Limviphuvadh[3], Akira Saito[1], Shunsuke Fujishiro[1], Yuichi Amemiya[1], Hiroyuki Toh[4] ([1]*Div. of Life Sci. & Eng., TDU, Japan*, [2]*MiB, Kyushu Univ., Japan*, [3]*BII, A*STAR, Singapore*, [4]*Dept. of Biomed. Chem., Sch. of Sci. & Tech., Kwansei Gakuin Univ., Japan*)

Many studies have reported that GPCRs function not only as their monomers but also as homo- or hetero-dimers or higher-order molecular complexes. Many GPCRs exert a wide variety of functions by a specific combination of GPCR subtypes. Besides, some GPCRs are reported to be associated with diseases. Thus, GPCR oligomerization is now recognized as an important event in various biological phenomena, and many researchers are investigating the subject. Recently, we have developed a high-performance method to predict interacting pairs for GPCR oligomerization by integrating structure and sequence information. In this session, we will discuss advantages and disadvantages of the method. In addition, we will introduce other attempts to investigate GPCR oligomers.

2SKA-06 1 分子イメージングで見る GPCR の多量体化とエンドサイトーシス
Single-molecule imaging of GPCR oligomerization followed by internalization

Masataka Yanagawa (*RIKEN*)

GPCR dimers have attracted much attention as potential drug targets because the dimerization regulates receptor functions including a ligand binding affinity, and G protein activation efficiency. However, a physiological importance of higher-order oligomerization of GPCRs is unclear, which cannot be distinguished from dimerization by conventional biochemical assays. Here we directly observed single-molecule dynamics of metabotropic glutamate receptor (mGluR), a prototypical class C GPCR, on the living cell surface by using total internal reflection fluorescence microscopy. The higher-order oligomerization of mGluR occurs with deceleration of the diffusion followed by the clathrin-dependent internalization, which is tightly coupled with receptor activation.

2SAP-01 熱揺らぎは分子モーターに共通する駆動力なのだろうか？
Is thermal fluctuation a common driving force of molecular motors?

Shuichi Nakamura (*Grad. Sch. Eng., Tohoku Univ.*)

Thermal fluctuation is a non-negligible external force for molecular motors working at nano/micro-meter scale. A thermal ratchet mechanism has been proposed in some molecular motors including kinesin, dynein and muscle myosin. We recently observed fast steps of the bacterial flagellar motor using a high-resolution nanophotometry and found that the step-speed distribution appears like a Boltzmann distribution. Our experiments provide the possibility that the flagellar motor is driven by thermal fluctuation, which might be a reason why flagellar motors are rotated even by the energy as low as k_BT. Does utilization of thermal fluctuation give molecular motors a high-efficiency operation mechanism?

2SAP-02 F_1-ATPase における異種エネルギー変換機構
Heterogeneous energy conversion mechanism of F_1-ATPase

Rikiya Watanabe[1,2] ([1]*Department of Applied Chemistry, The University of Tokyo*, [2]*PRESTO, JST*)

F_1-ATPase (F_1) is a rotary motor protein fueled by ATP hydrolysis. Compared to other motor proteins, F_1 is unique for its high efficiency and reversibility in converting chemical energy into mechanical work. In this presentation, I will introduce the recent findings on the rotary catalysis mechanism of F_1, i.e., chemo-mechanical energy conversion mechanism via stator-rotor interactions, which has been elucidated by in-house developed single-molecule manipulation techniques. The findings give us a clue for understanding how F_1 achieves the unique energy conversion mechanism among motor proteins.

2SAP-03 高速 AFM により導かれたミオシン V の力発生の原理
Principle for force generation in myosin V illustrated by high-speed AFM

Noriyuki Kodera[1,2], Takayuki Uchihashi[1,3,4], Toshio Ando[1,3,4] ([1]*Bio-AFM FRC, Inst. of Sci. & Eng., Kanazawa Univ.*, [2]*PRESTO, JST*, [3]*Div. of Math & Phys. Sci., Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [4]*CREST, JST*)

During high-speed AFM imaging, a controlled strong force can be applied onto a target locus of a molecule. With this technique, the trail head (T-head) of myosin V (M5) bound to actin in ATP-free solutions was mechanically detached. Upon every detachment, M5 stepped forward very similar to that seen in ATP, suggesting that the intramolecular tension for forward movement is generated without ATP energy. Once T-head is detached either mechanically or by ATP binding, the lead head swings forwards spontaneously. Thus, the ATP binding energy is mainly used for the movement. The role of ATP hydrolysis in M5 walking is only to ensure the unidirectional mechanical process, for which only ~$2k_BT$ is sufficient. This new view may hold also for other ATP-dependent bio-nanomachines.

2SAP-04 分子機械動作機構のクーロン的視点
Coulombic viewpoint of how molecular machines work

Mitsunori Takano (*Dept. Phys., Waseda Univ.*)

As we advanced our study on the allosteric mechanisms of molecular machines (from molecular motors to signal transduction proteins) using molecular dynamics simulation, we come to pay more attention to the Coulombic viewpoint of the mechanism. In this talk, I would like to discuss it by showing our recent results that made us think so.

2SAP-05 タンパク質分泌を駆動する反復モーターの作動原理
The working principle of repetitive motors driving protein export

Hiroyuki Mori (*Inst. Virus Res., Kyoto Univ.*)

The SecA ATPase and the SecYEG translocon play central roles in protein export across the bacterial cytoplasmic membrane. Membrane proteins SecDF are required for efficient protein export. Based on the structure-instructed biochemical analyses, it has been proposed that SecA functions as the ATP driven motor repeatedly pushing a secretory polypeptide into the SecYEG channel on the cytoplasmic side, and SecDF acts as the PMF-driven motor repeatedly pulling the translocating polypeptide on the other side of the membrane. In this symposium, I will introduce the working models and discuss how SecA and SecDF convert the ATP energy and the cation motive force energy, respectively, to the one-directional polypeptide movement.

2SAP-06 RNA ポリメラーゼの力発生機構
Force generation mechanism of the RNA polymerase

Yoshie Harada (*iCeMS, Kyoto University*)

RNA polymerase transcribes a DNA sequence, starting at a promoter site, into a chain of RNA. Search for the promoter involves linear diffusion of RNA polymerase along DNA, and nucleotide-dependent transcription along DNA is highly processive and accompanies a force as much as 25 pN. Because DNA is helical, one anticipates rotation of RNA polymerase around DNA during transcription and during the diffusional search. We showed that an RNA polymerase molecule attached to a glass surface rotated DNA for >100 revolutions around the right-handed screw axis of the double helix with a rotary torque of >5pN nm in 2001. Here, I will discuss about the mechanism of the force generation with a DNA motor particularly the RNA polymerase.

2SAP-07 １分子から階層を超えて細胞そして組織の機能へ繋ぐ
Bridging the hierarchy of single molecule and cellular and tissue functions

Toshio Yanagida[1,2] ([1]*RIKEN QBiC; NiCT*, [2]*Osaka Univ. CiNet*)

We have developed the single molecule detection technology for studying the mechanism of molecular motor and demonstrated that it does not overcome thermal noise (Brownian motion) but rather uses it for its function. This is in sharp contrast to the man-made machine. How is this nature advantageous for the function? In order to answer this question, we have used modeling and computer simulation, which indicates that the movement is due to a Brownian Ratchet mechanism and this mechanism successfully explains the adaptive force generation of muscle (cell). Furthermore, we have studied using the UT-heart simulator and 10-Peta Flops supercomputer “Kei” how the Brownian motion of individual molecular motors is successfully involved in the function of heart.

2SAP-08 疑おう
Let's doubt ourselves

Kazuhiko Kinosita, Jr. (*Dept. Phys., Fac. Sci. Eng., Waseda Univ.*)

If we stop doubting, we no longer do science. What do I doubt most? Myself. Thus I never cling to my own view (hopefully). What, then, should I tell you in this symposium? Facts? Well, facts, once described, are no longer objective truth. Perhaps I can ask questions, and then apologize that I cannot answer them.

2SAP-09 F1-ATPase が 100%近いエネルギー効率で回転する仕組み
A mechanism how F1-ATPase rotates with near 100% energy efficiency

Masasuke Yoshida, Ei-ichiro Saita (*Kyoto Sangyo Univ.*)

ATP synthase is a rotary motor enzyme and its catalytic portion F_1-ATPase hydrolyzes ATP to drive rotation of the central γ subunit. Efficiency of chemo-mechanical energy conversion by this motor is always near-perfect under different ATP hydrolysis energy by yet-unknown mechanism. We measured the torque as a function of rotation angle under different [ATP]/[ADP][Pi] and estimated mechanical work. The torque profiles show three sawtooth-like repeats during a net single ATP hydrolysis. When [ATP]/[ADP] and [P_i] in environment are changed, the height and hence the area (= mechanical work) of the sawtooth change accordingly so that mechanical work can always match available energy of ATP hydrolysis.

2SFP-01 ナノとマクロを繋ぐ生物電子顕微鏡アプローチ
Advanced electron microscopy: A new world view of mesoscale biology

Takuo Yasunaga (*Dept. of Biosci. and Bioinfo., Sch. of Comp. Sci. and Sys. Eng., Kyushu Inst. Tech.*)

Morphological understanding of meso-scale structures or organisms, such as cells and organelle, is essential for elucidation of their higher order functions of organisms from the macroscopic views. It is, conventionally, difficult to observe the three-dimensional structure at the nano-scale levels but advanced electron microscopy (EM) such as electron tomography and serial block-face SEM, makes it possible to observe them at the molecular level. Namely, we come to be able to obtain 3D finer structures/architecture in wider areas in situ / in vitro / in vivo under as native conditions as possible. Here, we introduce the recent progress of EM in cooperation with Japanese Society of Microscopy.

2SFP-02 クライオ電子線トモグラフィは細胞内タンパク質複合体構造解析に有力な手法である
Cryo-electron tomography is one of powerful techniques to understand the structures of intracellular protein complexes

Shinji Aramaki[1], Kouta Mayanagi[2], Kazuhiro Aoyama[3,4], Takuo Yasunaga[3,4] ([1]*Dept. of Bioscience and Bioinformatics, Kyushu Inst. of Tech.*, [2]*Medical Inst. of Bioregulation Kyushu Univ.*, [3]*FEI Company*, [4]*Research Center for Ultra-High Voltage EM, Osaka Univ.*)

Recently, a lot of protein structures were revealed at atomic resolution and accumulated to the databases. However, most proteins make complex network and realize their functions. Therefore it is important to understand the complex structures which have function in cells. In our study, we focus on filopodia of neuronal cells and tried to reveal molecular structures of cytoskeleton proteins networks with cryo-ET in order to understand the formational mechanisms of them. Then, we could get the detail of bundled actin filaments with bundling proteins and understand the architecture of filopodia. In addition to that, combination of the techniques and DDDs which are developed dramatically in this couple years will provide us novel knowledges about proteins in cells.

2SFP-03 電子顕微鏡によって明らかになったシヌクレインタンパク質の分子特性から細胞内輸送における役割を探る
Molecular properties of synuclein found through the electron microscopy and its function in the microtubule-based intracellular transport

Shiori Toba (*Graduate School of Medicine, Osaka City University*)

Synuclein is a one of microtubule binding proteins (MAPs) and a neuronal protein that is linked to Parkinson's disease. We combined electron microscopy of the recombinant synuclein protein and fluorescent microscopy of labelled synuclein in vivo and in vitro.
Electron microscopy studies demonstrated that synucleins facilitated tubulin polymerization under the critical concentration of microtubule formation (~1.1 mg/ml tubulin). Immunogold electron microscopy revealed that synucleins bound microtubules in a necklace-mimic fashion. Our findings through electron microscopy can give cues about the functional relevance of synuclein proteins, microtubules, and cytoplasmic dynein for the intracellular transport.

2SFP-04 低温電子線トモグラフィーにおける電子線直接検知型カメラの効果
The power of electron direct detector for electron cryotomography

Akihiro Kawamoto[1], V.Yusuke Morimoto[1,2], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ*, [2]*QBiC, RIKEN*)

Electron cryotomography (ECT) is known to be a powerful technique to visualize the three-dimensional (3D) structures of intracellular organelles in the whole cell. However, the contrast and resolution of 3D image obtained by ECT are limited because of radiation damage due to the requirement of recording many images for a tile series, and also intrinsically limited tilt angles cause blurring in one direction. Newly developed direct electron detector (DED) using CMOS-based sensors is dramatically improving image quality, allowing us to collect image data with better quality and much less radiation damage to attain higher resolution. I will present several examples of 3D image reconstruction using DED and discuss the power of DED for ECT.

2SFP-05 **Serial block-face SEM による細胞分裂の方向を決定する新規細胞膜構造の観察**

A novel plasma membrane structure to determine the orientation of the centrosome during cell division revealed by serial block-face SEM

Naoyuki Miyazaki[1], Takefumi Negishi[2], Naoto Ueno[2], Kazuyoshi Murata[1] ([1]*NIPS*, [2]*NIBB*)

Serial block-face SEM (SBF-SEM) is an advanced 3D electron microscopy technique for investigating large volumes at a resolution of a few tens of nanometers. In this method, thin surface of a resin-embedded specimen is cut off by a diamond knife attached to an in-chamber ultramicrotome, and then the newly exposed surface structure is imaged by SEM. The sectioning and imageing are automatically repeated to get a serial block-face images of the specimen. The 3D structure is reconstructed from the serial images after image alignment. Here, SBF-SEM observations revealed a novel plasma membrane structure to determine the orientation of the centrosome during cell division in the epithelial cells of ascidian, Ciona intestinalis embryo.

2SFP-06 **SEM ベース三次元再構築法（FIB-SEM トモグラフィー法）を用いたミトコンドリア-小胞体の相互関係の可視化**

3D organization of the mitochondria-associate membrane in mammalian cells by using FIB-SEM tomography

Keisuke Ohta[1,2] ([1]*Dept. Anatomy, Kurume Univ. Sch. Med.*, [2]*RIKEN, Qbic*)

Mitochondria-associated membranes (MAMs) are responsible for various cellular processes and recently have suggested to be involved in mitochondrial dynamics. However, the detailed 3D organization of MAM has not been established because there was no appropriate method until a few years ago. Recent advanced high-resolution focused ion-beam scanning electron microscopy (FIB-SEM) tomography is a unique method to visualize the organization of MAMs. In this method, 3D volume data is obtained by a cycle of FIB milling and SEM imaging. Resultant reconstruction data have a high depth resolution because each milling shave the specimen surface only a few nm per cycle, and the final resolution able to reach almost 10 nm, which is sufficient to analyze the organization of the MAM.

2SGP-01 **Sox2 の DNA 結合ドメインの構造揺らぎから分かった DNA 認識機構**

DNA binding mechanism of high-mobility group box domain of sox2 revealed by its conformational flexibility

Erisa Harada[1], Tsuyoshi Konuma[2], Shoko Mori[1], Kenji Sugase[3] ([1]*Suntory Foundation for Life Sciences*, [2]*Mount Sinai School of Medicine*, [3]*Kyoto University*)

Transcriptional factor protein sox2 possess a high-mobility group box (HMG) domain, which specifically binds to DNA to exert its function. To elucidate how sox2 recognizes the target sequence, we investigated dynamics of the HMG domain upon DNA binding using NMR. Although HMG domain is known to form three helices in the DNA complex, those helices are almost unfolded in the free state. Interestingly, structural change in the helical region was also observed when the HMG domain nonspecifically binds to random DNA sequences. This nonspecific DNA-binding complex shows conformational flexibility among multiple DNA-complex structures. These data suggest that the HMG domain adjusts its structure on DNA binding and finally forms the stable complex with the target sequence.

2SGP-02 **Dynamic Recognition of Unfolded Proteins by the Trigger Factor Chaperone as Investigated by NMR**

Tomohide Saio[1], Xiao Guan[2], Paolo Rossi[2], Charalampos Kalodimos[2] ([1]*Div. of Chem., Grad. School of Sci., Hokkaido Univ.*, [2]*CIPR, Rutgers University*)

Molecular chaperones prevent aggregation and misfolding of proteins and are thus central to maintaining protein homeostasis. However, scarcity of structural data has impeded an understanding of the recognition and anti-aggregation mechanisms of molecular chaperones.
Here we investigate the detailed mechanisms for recognition of unfolded substrate proteins by trigger factor (TF). Our results provide an insight into structure and dynamics of TF in complex with alkaline phosphatase (PhoA) as an unfolded substrate, showing how molecular chaperones prevent aggregation and promote the folding of substrate proteins.

2SGP-03 **Structural characterization of a denaturant denatured state**

Hironari Kamikubo (*MS, NAIST*)

Correct non-local contact formation during protein folding is an essential step to realize rapid formation of the native structure. In this study, in order to investigate a potential internal non-local collision in the denatured state, the lifetime measurement of tryptophan triplet state was applied to the denaturant denatured stated of staphylococcal nuclease. We prepared a series of Cys-substituted mutants on the amino acid residue arbitrary distant from W140. The triplet lifetimes of W140 with/without Cys were measured to estimate the quenching rate. In the results, the distribution of the quenching rates cannot be interpreted by assuming a random coil, and in addition exhibits frequent collision in the residues responsible for the native contacts.

2SGP-04 **天然物の結合によるヒトトランスサイレチンの構造変化**

Structural changes upon the binding of natural products in human transthyretin

Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

Transthyretin (TTR) is a serum protein associated with human hereditary amyloidosis. The discovery and development of small molecules that inhibit the amyloidogenesis of TTR is one of the therapeutic strategies for these diseases. Herein, we discovered that glabridin, ferulic acid phenethyl ester and γ-mangostin are the effective inhibitors against the amyloidogenesis of V30M amyloidogenic TTR by the limted screening, and the direct bindings to TTR were confirmed by *in-vitro* assays. Crystallographic analysis revealed the various binding modes. The binding of glabridin was associated with a induced-fit, whereas that of γ-mangostin was associated with the binding of two chloride ions. Structural changes upon the binding of inhibitors will be discussed in further detail.

2SGP-05 Protein-Protein interactions of the apicoplast proteins of Plasmodium falciparum

Takashi Saitoh[1], Shohei Yuasa[2], Fumina Oosaka[1], Katsumi Maenaka[1], Toshiharu Hase[2], Yoko Kimata-Ariga[2] ([1]*Grad. Sch. of Pharma. Sci., Hokkaido Univ.*, [2]*Inst. for Protein Research, Osaka Univ.*)

The plastid of Plasmodium falciparum, the apicoplast, of the malaria parasite is an essential organelle. The apicoplast is an organelle derived from a secondary endosymbiosis. Most apicoplast proteins are nuclear encoded and post-translationally targeted into the organelle using a bipartite N-terminal extension, consisting of a typical endomembrane signal peptide and a plant-like transit peptide. The malaria parasite possesses plant-type ferredoxin and ferredoxin-$NADP^+$ reductase in the apicoplast. We investigate protein-protein interactions in the apicoplast of Plasmodium falciparum based on structural biological and biophysical experiments. These interactions would be a promising drug target.

2SGP-06 リガンドとの相互作用に伴う膜蛋白質の動的立体構造変化
Dynamical structural change of membrane proteins upon interaction with their ligands

Takumi Ueda[1,2], Yutaka Kofuku[1], Ichio Shimada[1] ([1]*Grad. Sch. Pharm. Sci. the Univ. of Tokyo*, [2]*PRESTO, JST*)

Membrane proteins, which are embedded in the lipid bilayers of biomembranes, play various important roles in signal transduction, transportation, and bioenergetics, and over half of the currently available drugs target the membrane proteins. Recently, accumulating evidences have suggested that the membrane proteins are quite dynamic and interconvert between multiple conformations in equilibrium, and the population and exchange rates of the equilibrium determine their functions. I will talk about our NMR studies of the membrane proteins, which demonstrates that the ligands regulate the function, by changing the conformational equilibrium.

2SHP-01 プロトネーション状態の確認だけでなく機能探索の手法としての生体高分子中性子結晶学
Neutron protein crystallography as the technique for not only the identification of protonation state but also function investigation

Ichiro Tanaka[1,2] ([1]*Coll. of Eng., Ibaraki Univ.*, [2]*Frontier Res. Center*)

Protonation, hydration and hydrogen bond play important roles in various life processes at the atomic level. In order to study them, a new diffractometer for neutron protein crystallography (NPC), IBARAKI Biological Crystal Diffractometer (iBIX), has been constructed at BL03 in MLF in J-PARC and operational since December 2008. And J-PARC will arrive at 1MW proton maximum power at the target in the end of 2015 fiscal year. In this symposium, the possibility of the advanced investigation of function of proteins through hydrogen information will be discussed together with recent results by iBIX and other NPC instruments in the world.

2SHP-02 生体分子水和水の構造とカイネティクスの分子動力学解析
Atomic-Scale View of Biomolecular Hydration: From Structure to Kinetics

Yoshiteru Yonetani (*Japan Atomic Energy Agency, Quantum Beam Science Center*)

Precise structural information about biomolecular hydration has become available with recent high-resolution X-ray and neutron experiments. Although further information, kinetics of hydration water, is also important to understand the relevant dynamical processes, it is not yet fully clarified. One of the long-standing unsolved problems is the distinct timescale feature of hydration water. Some cases show a hydrogen-bond lifetime of a few pico-second, but in other cases it is over ~100 ps. What causes such a lifetime difference? Here, we carried out molecular dynamics simulations for various systems, DNA, protein, and ions in water, to explore the origin of the lifetime difference. Based on our results, we will also discuss our prospect of a future pKa study.

2SHP-03 タンパク質機能における内部結合水の役割
Role of bound water molecules inside proteins in their functional processes

Shigehiko Hayashi (*Grad. Sch. Sci., Kyoto Univ.*)

Bound water molecules inside proteins play a role in regulating reactivity of reaction sites through altering electrostatics around them, and in inducing large conformational changes of proteins coupled to change of chemical states of the reaction sites. Bound water molecules are highly controllable through small perturbation of electrostatics and conformational changes because of their nature of high mobility and polarity. The features are in turn exploited to amplify small triggering events to realize functional processes involving large chemical and conformational changes. In this talk, I present our recent studies on photoreceptor proteins by means of hybrid molecular simulations and discuss functional role of bound water molecules in their functional processes.

2SHP-04 膜チャネル・トランスポーターの分子機構
Molecular Mechanisms of Membrane Channel/Transporter

Osamu Nureki (*Department of Biological Science, Graduate School of Science, The University of Tokyo*)

Membrane channels and transporters mediate transport of the target solutes (ions, sugars, metabolites and xenobiotics) across the membrane, which is driven by chemical potential energy of the solutes themselves or partner ions. Recently, high-resolution structures of membrane proteins have been getting more and more available, due to the advancement of crystallization by lipidic cubic phase method, and to collect X-ray diffraction data from micro crystals using micro-focus beam in the synchrotron. These advancements allow us to fully understand the molecular mechanisms of membrane channels and transporters at an atomic resolution, which have uncovered that pKa of Glu and/or Asp residues largely affects their transport mechanism.

2SHP-05 ナトリウムポンプ型ロドプシンの機能におけるプロトンの役割

The role of proton on the function of sodium pump rhodopsin

Keiichi Inoue (*Nagoya Institute of Technology*)

Krokinobacter rhodopsin 2 (KR2) is a light-driven outward Na+ pump. It has a characteristic NDQ-motif in the third transmembrane helix consisting of N112, D116 and Q123. Although, NDQ-motif is highly conserved among Na+ pump rhodopsins, the mechanism of how these residues contribute to Na+-transport has not been revealed. In the recent study, we obtained new insights for the role of NDQ-motif. First, N112 forms transient Na+ binding pocket during the photocycle. On the other hand, Q123 optimizes the structure protein for the rapid uptake of Na+. Finally, it was revealed that H+ transfer occurs from protonated retinal Schiff-base to D116 upon the gate opening for Na+ uptake. This result indicates the transient change pKa balance plays crucial role for the Na+-transport.

2SHP-06 チトクロム酸化酵素の高分解能 X 線結晶構造解析から明らかとなった、酸素還元反応と共役したプロトン輸送機序

High-resolution X-ray structural analysis reveals how cytochrome c oxidase pumps protons coupled with molecular oxygen reduction

Atsuhiro Shimada[1], Naomine Yano[2], Kaori Kishida[3], Junpei Baba[1], Yuki Etoh[1], Keita Hatano[1], Eiki Yamashita[4], Kyoko Shinzawa-Itoh[1], Tomitake Tsukihara[1,4], Shinya Yoshikawa[1] ([1]*Picobiol. Inst., Grad. Sch. Life Sci., Univ. Hyogo*, [2]*Front. Res. Cent. Appl. Atomic Sci., Ibaraki Univ.*, [3]*Appl. Chem. in Biosci., Agrobio., Grad. Sch. Agri. Sci. Kobe Univ.*, [4]*Inst. Protein Res., Osaka Univ.*)

It has been proposed that cytochrome *c* oxidase (CcO) stores 4 protons from N-phase before sequential proton pump to P-phase. The proton pump is driven by pKa decrease in the proton pump pathway on electron transfer to the O_2-reduction site, although without showing the 4 proton capacity in the pathway. Our improvement of the resolution of X-ray structures of the fully oxidized/reduced CcOs from 1.8/1.9 Å to 1.5/1.6 Å reveals the existence of an isolated water cluster which reserves 4 pump protons and donates them actively to the pump pathway. Multiple conformers of D51 at the pump-pathway exit suggesting a significant proton affinity upon reduction are consistent to the reported proton-pump timing. These findings show a strongly ordered storage and release mechanism.

2SHP-07 Brownian ratchet mechanisms of macromolecular motors

Keiichi Namba (*Graduate School of Frontier Biosciences, Osaka University*)

Repeated association/dissociation of the rotor/stator drives step rotation of the bacterial flagellar motor, coupled with proton binding/release of an aspartate of stator proton channel. Distributions of step speed measured at sub-nm and near μs resolution by a nanophotometry system clearly show that the step motion is driven by thermal energy of Brownian motion. We also developed cryoEM image analysis methods to study actomyosin rigor complex structure and found how ADP and Pi are released from myosin upon its binding to F-actin and how myosin dissociates from actin filament upon ATP binding. Possible structure of weak binding state suggests directionally biased dissociation of myosin from actin filament, suggesting a Brownian ratchet mechanism of muscle contraction.

2SIP-01 シナプス分化を制御するスプライスインサート暗号の解読メカニズム

Decoding mechanisms of splice-insert signaling codes for synaptic differentiation

Shuya Fukai[1,2,3] ([1]*Life Sci. Div., SRRO, Univ. Tokyo*, [2]*IMCB, Univ. Tokyo*, [3]*JST CREST*)

Synapse formation is initiated by *trans*-synaptic interactions between adhesion molecules known as ‘synapse organizers’. Selection of synapse targets depends on selective pairing between pre- and postsynaptic organizers, which is regulated by splice inserts known as ‘splice-insert signaling codes’. Presynaptic type IIa receptor protein tyrosine phosphatases can induce synaptic differentiation through the splicing-dependent interaction with interleukin-1 receptor accessory protein (IL-1RAcP), IL-1RAcP-like 1 (IL1RAPL1) or Slit- and Trk-like family proteins. Based on their complex structures and structure-based mutational analyses, we reveal a structural basis for decoding of splice-insert signaling codes for synaptic differentiation.

2SIP-02 小脳シナプス形成を担う GluRδ2-Cbln1-neurexin 接着分子複合体の構造基盤

Structural insights into trans-synaptic GluRδ2-Cbln1-neurexin adhesion complex for cerebellar synapse formation

Takeshi Uemura[1,2] ([1]*Dept. Mol. Cell. Physiol., Inst. of Med., Acad. Assy., Shinshu Univ.*, [2]*CREST, JST*)

Synapse formation is triggered by interactions between trans-synaptic adhesion molecules. In the cerebellum, trans-synaptic interaction of postsynaptic glutamate receptor δ2 (GluRδ2/GluD2) and presynaptic neurexins (Nrxns) through cerebellin precursor protein 1 (Cbln1), a member of the C1q family, mediates synapse formation. Cbln1 is a ligand for both GluRδ2 and Nrxns and the binding of Cbln1 to Nrxns is tightly regulated by alternative splicing of Nrxns. Here, we present the crystal structures of the amino-terminal domain of GluRδ2 and Cbln1. By chemical cross-linking and MS analysis, we identified possible interaction sites of the complex. Based on these results, we propose a model of GluRδ2-Cbln1-Nrxns complex for cerebellar synapse formation.

2SIP-03 Solution behavior of TLR9 studied by analytical ultracentrifugation

Susumu Uchiyama (*Grad. Sch. Eng., Osaka Univ.*)

Sedimentation velocity analytical ultracentrifugation (SV-AUC) is a powerful biophysical method for studying protein-protein interactions. In this study, we investigated solution behavior of toll like receptor 9 (TLR9) in a wide range of concentration using SV-AUC with UV absorption system (ABS-SV-AUC) and fluorescence detection system (FDS-SV-AUC). Concentration dependent association of TLR9 with FITC-labeled DNA was monitored at nano-molar concentration range by FDS-SV-AUC, where TLR9 existed as a monomer even in the presence of the DNA. Whereas, monomer-dimer equilibrium of TLR9 was detected by ABS-SV-AUC at micro-molar concentration range in the presence of a specific DNA, providing a dissociation constant as 20 micro-molar for the monomer-dimer reaction.

2SIP-04 細胞表面受容体の結晶解析によって明らかになったシグナル伝達を制御する低親和性相互作用部位

Crystallographic analyses of cell-surface receptors revealed the presence of low-affinity interfaces regulating the signal transduction

Terukazu Nogi (*Grad. Sch. Med. Lif. Sci., Yokohama City Univ.*)

Cell-surface receptors are confined in the lipid bilayer and their motion is restricted to the two dimensional plane. Therefore, even low-affinity protein-protein interactions can trigger the signal transduction while it is quite hard to detect them by in vitro binding analyses using solubilized receptors or their partial fragments. X-ray crystallography, however, sometimes reproduces physiologically-relevant oligomeric assemblies or superstructures mediated by such weak interactions. In this talk, I would like to discuss the physiological significances of low-affinity assemblies discovered from the crystallographic analyses of cell-surface receptors implicated in development of the central nervous system.

2SIP-05 グルクロン酸糖結合レクチンとしてのクロトー共受容体

α-klotho is a high affinity lectin that binds terminal glucuronyl residues

Ryota Maeda[1,2] ([1]*Copenhagen Univ.*, [2]*IBRI*)

α-klotho (α-kl), a 130-kDa type I membrane protein, was first identified as an aging-related gene and later shown as a regulator of mineral homeostasis. Here, We determined the crystal structure of α-klotho at a 2.3 Å resolution, which is made of two domains structurally related to β-glycosidases, but without a catalytic machinery. Docking and molecular dynamics analyses show that the O-glycan of FGF-23 induces a conformational change in the α-Kl1 domain to form the hetero FGF-23/α-klotho complex. Altogether our findings demonstrate that α-klotho functions as a novel lectin capable of recognizing GlcA, including sulfated-GlcA-GalNAc and HNK-1-glycan, and provide insight into the physiological roles of glucuronylation in protein interactions.

2SIP-06 Toll 様受容体によるリガンド認識とシグナル伝達

Ligand recognition and signal transduction by Toll-like receptor

Umeharu Ohto (*Graduate School of Pharmaceutical Sciences, The University of Tokyo*)

Toll-like receptor (TLR), a family of the innate immune receptors, recognizes a wide range of microbial products and acts as a pathogen sensor. TLR is a type I transmembrane receptor consisting of an extracellular leucine-rich (LRR) domain responsible for ligand recognition, a transmembrane domain, and an intracellular TIR domain for signal transduction. Structural studies of TLR-ligand complexes have revealed molecular mechanisms by which each TLR specifically recognizes its own ligand. In this symposium, I will discuss the similarities and diversities of TLR-ligand interactions and signaling mechanisms.

3SAA-01 繊毛・鞭毛の 96 nm 周期を決める分子ものさし

A molecular ruler determines the repeat length in eukaryotic cilia and flagella

Masahide Kikkawa (*The University of Tokyo*)

Existence of cellular structures with specific size raises a fundamental question in biology: How do cells measure length? One conceptual answer to this question is by a molecular ruler, but examples of such rulers in eukaryotes are lacking.
Here, we studied the FAP59/172 complex by using genetics and cryo-electron tomography (Cryo-ET). The two proteins form a complex, and their absence disrupts 96-nm repeats in axonemes. Cryo-ET revealed that the FAP59/172 complex takes a 96-nm-long extended conformation along axonemal microtubules. Elongation of the complex resulted in extension of the repeats and duplication of specific axonemal components. We conclude that the FAP59/172 complex is the molecular ruler that defines 96-nm repeats in cilia/flagella.

3SAA-02 細胞膜へのフォスファチジルセリンの暴露を制御するフリッパーゼとスクランブラーゼ

Flippase and scramblase that regulate the phosphatidylserine-exposure to plasma membrane

Shigekazu Nagata (*Immunology Frontier Research Center, Osaka University*)

Two membrane proteins (TMEM16F and Xkr8) are involved in scrambling of phospholipids in plasma membrane. TMEM16F carries 8 transmembrane regions, requires Ca2+ to mediate phospholipid scrambling, and plays a role in the PtdSer-exposure in activated platelets. Xkr8 carries 6 transmembrane regions, and Caspase 3 cleaves off the C-terminal tail of Xkr8 to execute its scramblase activity in apoptotic cells. In addition to the activation of scramblase, the flippase that translocates PtdSer from outer to inner leaflets is inactivated during apoptosis. We found that ATP11C of a P4-type ATPase together with its chaperone CDC50A works as a flippase, and is inactivated by caspase during apoptosis. Here, I discuss on the molecular mechanism of scramblase and flippase.

3SAA-03 CRISPR-Cas9 の結晶構造

Crystal structure of CRISPR-Cas9

Hiroshi Nishimasu (*The University of Tokyo*)

Cas9 is an RNA-guided DNA endonuclease and is implicated in the bacterial CRISPR-Cas adaptive immune system. Cas9 binds guide RNA, and cleaves target double-stranded DNA complementary to the guide RNA. Since Cas9 can induce site-specific DNA double-strand breaks in the genome in various cell types, it has been attracting a lot of attention as a new, versatile genome-editing technology. To understand the RNA-guided DNA targeting mechanism, we solved the crystal structures of Cas9 in complex with guide RNA and target DNA. The complex structures provided insights into the RNA-guided DNA targeting mechanism of Cas9, and enabled the rational design of new, Cas9-based tools.

3SAA-04 25 サブユニットからなる転写メディエーター複合体の再構成

Total reconstitution of the 25-subunit Mediator complex of transcription regulation

Tsuyoshi Imasaki[1,2,3], Papi Gabor[2,4], Kentaro Yamada[2], Schultz Patrick[4], Yuichiro Takagi[2] ([1]*JST researcher*, [2]*Indiana Univesity*, [3]*RIKEN*, [4]*IGBMC*)

Mediator is the large multi-protein complex composed of 25 subunits with molecular mass over 1 mega daltons. Mediator regulates RNA polymerase II transcription in eukaryotes. The size, complexity, low abundance of Mediator has severely compromised structural-functional studies over the years. By utilizing our latest protein complex expression technology, the 25-subunit entire Mediator complex has been successfully reconstituted, opening a door for rigorous structure and functional studies for the first time, leading toward high-resolution structure determination of the entire complex by X-ray crystallography.

3SAA-05 オートファジーの始動を担う Atg1/ULK 複合体の構造と機能

Structure and function of the autophagy initiating Atg1/ULK complex

Nobuo N. Noda[1,2] ([1]*Inst. Microbial Chem.*, [2]*JST, CREST*)

In autophagy, autophagosomes sequester a portion of cytoplasm and deliver it to the lysosome/vacuole for degradation. Autophagosome formation requires 18 Atg proteins, which have been classified into six functional groups. The Atg1/ULK complex is one of the six groups and functions as the most upstream factor among them. Yeast Atg1 complex consists of Atg1, Atg13, Atg17, Atg29 and Atg31, whereas its mammalian counterpart ULK complex consists of ULK1/2, Atg13, FIP200 and Atg101. We determined the crystal structures of their sub-complexes and established structural basis of the Atg1/ULK complex. We are now trying to unveil the overall architecture and molecular functions of this complex in autophagy initiation using several techniques including high-speed AFM.

3SBA-01 Hybrid Approaches to Characterize Structure and Dynamics of Biomolecular Systems from Single Molecule Experiments

Florence Tama[1,2] ([1]*Nagoya University, Physics*, [2]*RIKEN AICS*)

Low-resolution experimental techniques such as cryo-EM are often used to characterize structure of biomolecules. X-ray free electron laser (XFEL) is an also exciting new technology that could significantly extend our structural knowledge of biological molecules. We will present computational methods to obtain atomic level descriptions of conformational transitions occurring in macromolecular complexes using a variety of low-resolution experiments and X-ray crystallography. We will also present a new method, which compares electron microscopy 2D images with computationally predicted motions of biomolecules, therefore, effectively identifying motions taking place in the protein sample.

3SBA-02 2D hybrid analysis: An approach to build 3D atomic model from 2D EM image

Atsushi Matsumoto (*Japan Atomic Energy Agency*)

We have developed a computational approach to build an atomic model from an electron microscopy (EM) image of a biological molecule. In this approach, many atomic models of the molecule with different conformations are prepared first by deforming the X-ray crystal structure or the modeled structure using a computational technique. Then, a variety of orientations is given to each atomic model to obtain projection images. Finally, the projection images are compared to the EM image. The atomic models with the projection similar to the EM image are regarded as the candidates for the atomic structure of the molecule. In the symposium, the application to the giant cadherin proteins, which are involved in the cell adhesion, will be explained.

3SBA-03 分子動力学シミュレーションと電子顕微鏡像を用いたハイブリットシミュレーションによるリボソーム内 tRNA 転位の解析

Analysis of tRNA translocation through the ribosome by a hybrid-simulation using an MD simulation and electron microscopy density maps

Hisashi Ishida (*Japan Atomic Energy Agency, Quantum Beam Science Center, Molecular Modeling and Simulation Group*)

To understand the transition path between different reaction states and the free-energy profile along the path, a hybrid-simulation using all-atom molecular dynamics (MD) simulations and electron microscopy (EM) density maps was developed. The hybrid-simulation comprises two stages; one is to predict a transition path starting from an X-ray structure to EM density maps using an all-atom MD simulation in water medium. The other is to sample conformations to obtain the free-energy profile along the predicted path. The hybrid-simulation was applied to the system of ribosome-tRNAs-EFG to understand the mechanism of tRNA translocation. The results showed that a ratche-like motion, the movement of the P/E-gate and EF-G play important roles in tRNA translocation.

3SBA-04 X 線溶液散乱と二次構造情報によるタンパク質の立体構造の構築

Construction of Protein Structure by Small-Angle X-ray Scattering Constraints and Secondary Structural Information

Masaki Kojima, Yasumasa Morimoto, Takayuki Ichioka (*Tokyo University of Pharmacy and Life Sciences*)

Using a hybrid approach with small-angle X-ray scattering (SAXS) and restrained molecular dynamics, we developed a new method to construct protein structures by SAXS constraints and secondary structural information. Our method calculates the constraint force derived from the differences between observed and calculated SAXS intensities, and applies it to each atom so that the resultant structure could satisfy the experimental SAXS data. We performed the calculation for nine proteins with different folds using SAXS constraints and NMR-derived distance restraints for secondary structures. Based on these results, we were successfully able to construct coarse-grained molecular models of these proteins at amino acid residue resolution.

3SBA-05 様々な顕微鏡法によるアクチンフィラメント構造解析
Structural analysis of the actin filament by several microscopy techniques

Akihiro Narita[1,2] ([1]*Struct. Biol. Res. Center, Nagoya Univ.*, [2]*JST PRESTO*)

Actin is one of most abundant proteins, which forms a double stranded filament. Actin continuously polymerizes and depolymerizes in the cell and this dynamics is crucial for many important phenomena in the cell. We are investigating the dynamics of the actin filament by structural analysis using several techniques including cryo-electron microscopy, negatively staining, single particle analysis, electron tomography, STEM and AFM. Each technique has clear merits and demerits and it is useful to select suitable method for each purpose. For example, STEM showed large signal to noise ratio without CTF deformation for negatively stained sample. I would like to report comparison between the techniques with real data and discuss how to select them.

3SCA-01 流動電位測定により明らかにされたカリウムチャネルポア内でのイオンと水の共役的透過
The flux coupling between ion and water in the narrow pore of the K^+ channels revealed by the streaming potential measurement

Masayuki Iwamoto, Shigetoshi Oiki (*Dept. Mol. Physiol. Biophys., Univ. Fukui Facult. Med. Sci.*)

The crystal structure of potassium channels revealed single-filed ion distributions in the narrow pore (the selectivity filter). Water molecules are intercalated between ions in the selectivity filter, thus, water is driven to flow upon ion permeation. This coupling of water and ion fluxes (J) underlies permeation process of ion channels. The water-ion coupling ratio (J_w/J_i) can be quantitatively evaluated by electrophysiologically measured streaming potentials (V_{stream}), which is based on the principle of the non-equilibrium thermodynamics. We developed the method and showed that the J_w/J_i value increases substantially as the K^+ concentration decreases. Quantitatively evaluated water fluxes serve for elucidating the permeation processes through the channel.

3SCA-02 電気生理で膜輸送体の ATP 加水分解メカニズムを解明する
Monitoring ATP-hydrolysis cycle by electro-physiological approach: Patch-clamp recordings of CFTR

Yoshiro Sohma (*Keio Univ. Sch. Med.*)

Cystic fibrosis transmembrane conductance regulator (CFTR) channel is a unique member of the ABC transporter superfamily which shares two Nucleoside Binding Domains (NBD), because most of the ABC members function as a solute transporter.
In CFTR, the ATP-driven 'NBD engine' drives the channel gate in Membrane Spanning Domain (MSD), which allows us to investigate the mechanism of the NBD engine by patch-clamp technique. By combined use of channel current recordings and gating simulation, we elucidated the dynamism of NBD engine during ATP-hydrolysis cycle and the coupling between the ATP-hydrolysis cycle at NBD and the gating cycle at MSD.
In this symposium, I will introduce application of the patch-clamp technique for the CFTR function-structure relationship study.

3SCA-03 オメガ-3 脂肪酸は Slo1 BK チャネルを活性化する
Omega-3 fatty acids activate Slo1 BK channels

Nobuyoshi Tajima[1], Yutao Tian[2], Rong Xu[2], Stefan Heinemann[3], Shangwei Hou[4], Toshinori Hoshi[2] ([1]*Dep. Physiol., Kanazawa Medical University*, [2]*Dep. Physiol., University of Pennsylvania*, [3]*Center for Molecular Biomedicine, Dep. Biophys., Friedrich Schiller University Jena*, [4]*Shanghai Center for Systems Biomedicine, Shanghai Jiao Tong University*)

Large-conductance Ca^{2+}- and voltage-dependent K^+ (Slo1 BK) channels are widely expressed in excitable and non-excitable cells of mammals. In vascular smooth muscle cells, BK channels provide a vasodilatory influence. We found that Long-chain omega-3 fatty acids such as docosahexaenoic acid (DHA) with EC_{50} of ~500 nM directly and reversibly activates BK channels composed of the pore-forming Slo1 subunit and the auxiliary subunit β1 in excised-patches, increasing currents by up to ~20-fold. DHA ethyl ester (DHA EE), found in dietary supplements, fails to activate BK channels and antagonizes the stimulatory effect of DHA. In this symposium, I will show electrophysiological approaches that were used to find the results mentioned above.

3SCA-04 破骨細胞における細胞膜 V-ATPase 電流の経時的解析
Real-time analysis of V-ATPase (proton pump) currents in the plasma membrane of osteoclasts

Miyuki Kuno (*Dept. Physiol., Osaka City Univ. Grad. Sch. Med.*)

Vacuolar proton ATPases (V-ATPases) are one of most ubiquitous mechanisms responsible for generating acidic environments in both intracellular and extracellular spaces. The uphill H+ transport by the V-ATPase utilizes energy produced by ATP-hydrolysis. However, the activity of the V-ATPase can be regulated in various ways, and the H+ transport is dependent on both pH- and voltage-gradients across the membrane. As a consequence of the diversity of the regulatory mechanisms, the ability of the pump to transport H+ could vary under physiological/pathological cellular conditions. We used whole cell clamp recordings to identify the pump currents in the plasma membrane of osteoclasts, and evaluated the changes during exposure to high concentrations of Ca2+.

3SCA-05 電位依存性 H^+チャネル動的構造への電気生理学的アプローチ
Electrophysiological approaches to structural dynamics of the voltage-gated H^+ channel

Yuichiro Fujiwara (*Grad. Sch. of Med., Osaka Univ.*)

The voltage-gated H^+ channel (Hv) is a H^+-permeable four-transmembrane domain protein that corresponds to the voltage-sensor domain of other ion channels and phosphatases. The functional unit of Hv is a homo dimer assembled by the cytoplasmic coiled-coil domain. The structural mechanism how the dimerization controls the gating and how Hv senses the membrane potential have been unknown. I have tried to dissect the structural rearrangement of the transmembrane helices associated with the channel gating. In this symposium, I will introduce electrophysiological approaches to the structural dynamics of Hv that revealed that a pair of voltage-sensor helices within the dimer works as a single unit according to the gating process.

3SCA-06 蛍光を使ってイオンチャネルの電流と電位センサーの動きを同時に測定する

Simultaneous recordings of ionic currents and voltage sensor movements by voltage clamp fluorometry

Koichi Nakajo (*Dept. Physiol., Osaka Med. Coll.*)

Electrophysiological techniques are very powerful tools to study ion channels and electrogenic pumps/transporters. However, the ionic currents are basically the final output of the gating process (i.e. opening of the gate), therefore, non-electrogenic process or conformational changes cannot be directly detected by electrophysiological methods. The voltage clamp-fluorometry (VCF) has been used to detect the conformational changes of membrane proteins using fluorophore covalently attached to the domain of interest. I will introduce some examples of VCF including KCNQ1 potassium channel, whose gating is dramatically slowed down by the accessory subunit KCNE1. The VCF revealed that KCNE1 hampers the coupling between the voltage sensor and the gate of KCNQ1 channel.

3SDA-01 胃プロトンポンプのアンタゴニスト結合構造

Antagonist-bound structures of gastric proton pump

Kazuhiro Abe[1,2] ([1]*CeSPI, Nagoya Univ.*, [2]*Grad. Sch. Pham., Nagoya Univ.*)

Gastric H,K-ATPase, an ATP-driven proton pump responsible for gastric acidification is a molecular target for acid suppressants. Systematic comparison of the molecular conformation of several 3D structures of H,K-ATPase, with or without bound antagonist, revealed that the A-M2 linker importantly contributes to conformational changes of the enzyme, especially those required for the luminal gating.
We also determined 7Å structure with bound BYK99, a potent antagonist of H,K-ATPase compared with previously applied SCH28080. Using homology model, docking simulation, and mutagenesis, we propose a binding model of H,K-ATPase antagonist which is a prototype of currently developing acid suppressants for the treatment of gastric acid-related diseases.

3SDA-02 Ca^{2+}-ATPase の触媒部位と輸送部位間の M2 ヘリックスを介したロングレンジ共役

Long-range Coupling between Catalytic and Transport Sites via Second Transmembrane Helix (M2) in Ca^{2+}-ATPase

Takashi Daiho, Kazuo Yamasaki, Stefania Danko, Hiroshi Suzuki (*Biochem., Asahikawa Med. Univ.*)

Ca^{2+}-ATPase couples ATP hydrolysis with Ca^{2+} transport, and forms an obligatory phosphorylated intermediate. The large rotational movements of Actuator (A) domain rearrange the distant transport sites through connections with transmembrane helices M1 and M2. Here we explore the structural roles of M2 in the coupling and in each of intermediate processing stages by mutationally disrupting helix and elongating at each of its cytoplasm and transmembrane regions of M2 and its junction with the A domain. Results pinpoint which parts of M2 are critical for cytoplasm or luminal gating at each stage, and suggest that proper gating requires appropriate interactions, tension and/or rigidity in the M2 regions at adequate times for coupling with A-domain movements and catalysis.

3SDA-03 Na+,K+-ATPase は膜貫通結合部位に結合した K を順番に置換する- X 線結晶解析を用いたキネティックス測定

Sequential substitution of bound K+ in the transmembrane binding sites of Na+,K+-ATPase, Kinetics by X-ray crystallography

Haruo Ogawa[1], Ayami Hirata[1], Flemming Cornelius[2], Chikashi Toyoshima[1] ([1]*IMCB, The University of Tokyo*, [2]*Department of Biomedicine, Aarhus University*)

Na+,K+-ATPase is one of the most important members of the P-type ATPase family. It transports 3 Na+ from the cytoplasm into the extracellular medium and 2 K+ in the opposite direction per ATP hydrolyzed. The binding and release of Na+ and K+ occur all sequentially. We demonstrate by X-ray crystallography of the ATPase in a state analogous to E2.Pi.2K+, combined with isotopic measurements, that the substitution of the 2 K+ with congeners in the extracellular medium occurs faster at site II. An analysis of B-factors of protein atoms in the crystal shows that the M3-M4E helix pair opens and closes the ion pathway leading to the extracellular medium. These results indicate that site I K+ is the first cation to bind to the empty cation binding sites after releasing 3 Na+.

3SDA-04 リン脂質 flippase である P4-ATPase の基質特異性と細胞機能の関係

Phospholipid flippase activities and substrate specificities of P4-ATPases and their roles in cellular function

Hye-Won Shin (*Grad. Sch. Pharm. Sci., Kyoto Univ.*)

The lipid bilayer of the plasma membrane and organelle membranes exhibits asymmetric lipid distributions. The spatiotemporal changes of lipid compositions between the bilayers are closely related to a variety of cellular functions. P4-ATPases, a subfamily of P-type ATPases, translocate phospholipids from the exoplasmic to the cytoplasmic leaflet of cellular membranes. We determined the subcellular localization of 13 human P4-ATPases and revealed the flippase activities and substrate specificities of the plasma membrane localized P4-ATPases. We found that an enhanced substrate-specific flippase activity is associated with the plasma membrane dynamics. We also showed the flippase activity of ATP8B1 is associated with the episode of genetic cholestatic liver disease.

3SDA-05 マラリア原虫のクロロキン耐性トランスポーター(PfCRT)の生成再構成系を用いた機能解析

Functional analysis of Plasmodium falciparum chloroquine resistance transporter (PfCRT) by reconstituted system with purified protein

Hiroshi Omote[1], Narinobu Juge[2], Sawako Moriyama[1], Takaaki Miyaji[2], Mamiyo Kawakami[1], Haruka Iwai[1], Tomoya Fukui[1], Yoshinori Moriyama[1] ([1]*Dept. of Membrane Biochemistry, Okayama University*, [2]*Adv. Science Research Center, Okayama University*)

Plasmodium falciparum CQ resistance transporter (PfCRT) plays essential role in CQ resistance of the malaria parasite. PfCRT having CQ resistant mutations is believed to export CQ from digestive vacuole. However, the physiological significance and biochemical properties of PfCRT have not been known. We have analyzed PfCRT using reconstituted system. PfCRT was over-expressed in the bacteria and purified through Ni-NTA column chromatography. Reconstituted PfCRT transported tetraethylammonium, verapamil, CQ, basic amino acids, polypeptides, and polyamines in a H+ symport manner. Furthermore, CQ competitively inhibited amino acids transport. These results indicated that PfCRT functions as a H+-coupled polyspecific nutrient and drug exporter.

3SDA-06 集合に共役した細菌べん毛モーター固定子ユニットの活性化機構

Assembly-coupled activation of the torque-generating stator units in the bacterial flagellar motor

Seiji Kojima (*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*)

Torque of the bacterial flagellar motor is generated by the rotor-stator interaction that couples with the ion flow through the channel in the stator. A dozen of stator units surround a rotor, and they must be anchored through a peptidoglycan-binding domain in the periplasmic region of the stator B subunit. Furthermore, ion-conducting activity of the stator units are activated only when they are incorporated into the motor. To unravel how active stator units function in the motor, we performed structural analyses of the soluble parts essential for function in the stator B subunit. Following structure-guided functional analyses provided insightful results for proposing the assembly-coupled activation mechanism of stator units, which we will discuss in the meeting.

3SDA-07 膜輸送蛋白等の計測における１分子顕微鏡観察とマイクロデバイスの活用

Single-molecule observation and utilization of microdevice for measurements of membrane transport proteins

Tomoko Masaike[1,2,3] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Science*, [2]*Res. Inst. for Sci. and Tech., Tokyo Univ. of Science*, [3]*PRESTO, JST*)

Single-molecule and microdevice techniques have recently become keys to pursuing studies of membrane transport proteins. Minimal functional units are labeled with fluorescent probes and observed for investigation of unitary steps in chemical reactions or structural dynamics of single molecules. We introduce application of such strategies to an ion pump Ca^{2+}-ATPase. Meanwhile, utilization of microdevice revolutionizes measurements of membrane proteins. It is exemplified by simple formation of lipid bilayers and increased S/N ratio in electrical measurements of gramicidin A using parylene double well chip. Apart from membrane proteins, expanded application of these techniques to ciliary motility, dynamics of microtubules, and detection of P_i are also illustrated.

3SFA-01 微生物に含まれる脂質の構造解析のための MS-AFM-IR システム

MS-AFM-IR platform for structural analysis of lipid inclusions in micro-organisms

Eric Lesniewska, P. Vitry, A. Dazzi, M.−J. Virolle, L. Tetard, E. Bourillot (*ICB UMR CNRS 6303, Univ. of Bourgogne Franche-Comte*)

Identification of entities and lipid inclusion inside cells with non-destructive techniques is a challenging procedure. We propose to couple the chemical characterization by IR spectroscopy with acoustic microscopy used for tomographic reconstruction [1-2]. The present study focuses on Streptomyces bacteria that can store its carbon source into TriAcylGlycerols, a potential direct source of biofuel [3]. These bacteria were imaged with this new platform and highlight the presence of lipid vesicles not accessible by classical topographical techniques.

[1] Vitry et al. Nanoscale 12274, 2015.
[2] Tetard et al. Nature nanotechnology, 3(8), 501-505, 2015.
[3] Deniset-Besseau et al. J. Phys. Chem. Lett., 5(4), 654-658, 2014.

3SFA-02 原子間力顕微鏡：細胞力学特性の個性を測る

Atomic force microscopy: Quantifying mechanical variation in living cell system

Takaharu Okajima (*Grad. Sch. Inform. Sci. Tech., Hokkaido Univ.*)

Cells have a large intrinsic spatial and temporal variation in cell functions. We notably focus on understanding the origin of cell mechanical variability, which is crucial for cells to maintain their ability in environments. Here, we describe our recent progress in understanding such a variation at the single cell level and in cell population observed by atomic force microscopy (AFM). The AFM combined with micro-fabricated substrates revealed that the cell-to-cell variation in cell modulus exhibits a characteristic frequency dependence that follows a single power-law rheology and is consistent with the temporal variation, satisfying an ergodicity in living cell system. The AFM also allows us to unveil emerging large-scale mechanical properties of cell population.

3SFA-03 新しい熱プローブを用いた細胞発熱計測および分子モーター活性制御

New thermal probes for cellular heat measurement and for temporal regulation of motor proteins

Yuichi Inoue (*IMRAM, Tohoku Univ.*)

Understanding heat transfer at cellular level or single molecular level is a frontier in biophysics. With a view toward examining the thermal environment of biomolecules, we developed two types of the thermal probes. The first probe is a bimaterial microcantilever to detect local heat generation. The second probe is single carbon nanotube as a new platform for motor proteins to induce local temperature change. The results of temperature measurements and estimations of thermal gradient with finite element method will be presented to discuss about future applications to measure and/or manipulate thermal environment of around biomolecules.

3SFA-04 細胞構造と機能を操る高圧力顕微鏡法

High-pressure microscopy for manipulating cellular architecture and function

Masayoshi Nishiyama (*The HAKUBI Center, Kyoto Univ.*)

Water is the most abundant substance in cells. Most intracellular reactions occur with the association of water molecules that surround the protein molecules. Application of pressure is a powerful method for modulating intermolecular interactions between protein and water molecules. This means that applied pressure is able to modulate the structure and function of protein molecules, without requiring the use of any chemical materials other than water molecules. We have developed a microscope that enables us to acquire high-resolution microscopic images at high-pressure conditions. The developed system allows us to visualize and manipulate the cellular architecture and activity.

3SFA-05 集光レーザー摂動を用いた神経回路網における分子ダイナミクスの直接操作

Direct manipulation of molecular dynamics in neuronal network with laser-induced perturbation

Chie Hosokawa (*Biomed. Res. Inst., AIST*)

In order to realize artificial control of functional connectivity in cellular network without any drugs and genes, it is indispensable to develop novel approaches using spatially and temporally controlled perturbation method into single cells. Here, we demonstrate laser-induced perturbation into molecular dynamics with optical tweezers. The optical trapping and assembling dynamics of synaptic vesicles, neural cell adhesion molecules, and receptors labeled with quantum-dot on neuronal cells were investigated by fluorescence analysis. In addition, we demonstrate enhancement of optical trapping force induced into molecules using non-resonance and resonance laser beams.

3SFA-06 *Xenopus* 卵抽出液を封入した小胞中でのアクチンの流れと小胞運動

Actin flows in *Xenopus* egg extract confined in oil and generates a force for migration of the extract

Naoki Noda, Issei Mabuchi (*Grad. Sc. Sci., Gakushuin Univ.*)

Actin dynamics in *Xenopus* egg extract confined in oil and surrounded by a phospholipid membrane was investigated. In the "extract droplet", an aggregate of cytoplasmic materials, which we call *X* body, was formed and actin flowed in a centripetal manner toward the *X* body for about an hour. The flow initiated at the oil-extract interface and terminated at the *X* body but the actin did not accumulate at the *X* body. The speed of the actin flow was decreased by addition of a myosin ATPase inhibitor, blebbistatin. Furthermore, the flow could induce migration of the droplet on substratum in the direction opposite to that of the flow. The force generated in the migrating droplet was similar to that generated by moving keratocyte.

3SGA-01 自己集積的に形成するキネシン・微小管の収縮性ネットワーク：工学応用に向けて

Self-organized Contractile Networks of Microtubules and Engineered Kinesins: Towards Engineering Applications

Yuichi Hiratsuka[1], Takahiro Nitta[2] ([1]*Sch. Mat. Sci., JAIST*, [2]*Appl. Phys. Course, Gifu Univ.*)

In living cells, motor proteins and cytoskeletons are ubiquitous, and involved in various cellular functions. The functions can be achieved through distinct self-organized subcellular structures, such as contractile rings and stress fibers. Inspired by this, we intended to develop a molecular system consisting of microtubules and kinesins which can self-organize into various morphologies and to use it for microdevices. By using a genetically engineered kinesin which can form loosely coupled dimers, we found that microtubules and the kinesins were self-organized into contractile networks. The networks took mesh-like and fiber-like morphologies, depending on structures of microchambers in which the networks were assembled. We will show demonstrations on the applications.

3SGA-02 Formation and rupture of a motorized cytoskeletal network

Takayuki Torisawa[1,2], Daisuke Taniguchi[2,3] ([1]*Advanced ICT Research Institute, NICT*, [2]*CREST, JST*, [3]*Dept. of Physics, School of Science and Technology, Meiji Univ.*)

We reconstructed simple microtubule-kinesin systems which yield various spatiotemporal dynamic. The static network, active network, aggregation phase, and population of asters emerged according to the concentrations of the system components and the motor activities. A coarse-grained model revealed two determinative features for the formation of these spatiotemporal patterns: First, motors must accumulate to form a number of interconnected spots. Second, the magnitude of the force between spots must nonlinearly depend on motor concentration. We also found that energy is stored and can be transformed into mechanical work during the rupturing dynamics of active network. This mechanical feature might contribute to symmetry breaking in cell locomotion.

3SGA-03 神経幹細胞のネマチックパターン

Active nematics of collective neural stem cells

Kyogo Kawaguchi[1], Ryoichiro Kageyama[2], Masaki Sano[3] ([1]*Dept. Syst. Biol., Harvard Med. School*, [2]*Inst. for Virus Res., Kyoto Univ.*, [3]*Dept. Phys., Univ. Tokyo*)

Neural stem (NS) cells are an ideal system to quantitatively measure fundamental single-cell behaviors underlying neurogenesis. Resembling its feature in vivo, monolayer-cultured NS cells show characteristic bipolar morphology and motion in the direction of their axes. Here we report the observation of dynamic liquid crystal-like pattern formed by NS cells. The nematic interaction between single cells induce local alignment and unstable topological defects in the two-dimensional geometry.

3SGA-04 Inference for the mechanics of moving cell sheets

Yohei Kondo[1], Kazuhiro Aoki[2], Shin Ishii[1] ([1]*Grad. Sch. Info., Kyoto Univ.*, [2]*Grad. Sch. Med., Kyoto Univ.*)

Mechanical forces generated by the cells drive morphogenetic processes of living tissues during development and regeneration, and recent advances in experimental and theoretical techniques enabled us to quantify the forces. However, there has not been thorough studies on how the tissues deform and flow under the forces, and we are still far from quantitatively understanding the dynamics of tissue shape. Here we are trying to develop a statistical method to mathematically approximate the dynamics at a macroscopic level. Our current strategy is to interpret the tissue as a continuum mechanical system, and use the observed velocity and force fields to compute the maximum likelihood estimate of the model parameters.

3SGA-05 線虫 C. elegans における減数分裂期細胞質流動の自己組織化の機構

A mechanism of self-organization in meiotic cytoplasmic streaming of the C. elegans embryo

Kenji Kimura[1,2], **Akatsuki Kimura**[1,2] ([1]*Cell Arch. Lab., Nat. Inst. Genetics*, [2]*Dept. Genetics, SOKENDAI*)

Cytoplasmic streaming is a collective movement of intracellular materials. In most cases, the direction of the flow is pre-determined due to the polarity of the cell. In contrast, the direction of a meiotic cytoplasmic streaming in the worm, C. elegans embryo is unlikely pre-determined. Here we address how the activities of individual motor protein are organized into the cell-wide collective streaming. We conducted live cell imaging combined with gene knockdown analyses and image processing to characterize the streaming quantitatively. We also succeeded to construct a numerical model that recapitulated important features of the streaming. This cytoplasmic streaming offers a new example of spatiotemporal pattern formation in biological systems.

3SHA-01 インフルエンザウイルス感染と宿主細胞侵入時に惹起される細胞内シグナルの可視化

Visualisation of molecular events during influenza virus entry and infection

Yusuke Ohba[1], Yoichiro Fujioka[1], Kazuhito V. Tabata[2], Shinya Nishide[2], Asuka Nanbo[1], Hiroyuki Noji[2] ([1]*Hokkaido Univ. Grad. Sch. Med.*, [2]*Grad. Sch. Eng., Univ. Tokyo*)

Influenza A virus (IAV) infection causes highly contagious, severe respiratory disorders, leading to thousands of deaths every year; however, the molecular mechanism for the IAV transmission pathway before replication has yet to be elucidated. We have reported that phosphoinositide-3-kinase (PI3K) bound to activated Ras in the endosomes, which participates in the regulation of IAV entry into host cells. Furthermore, signalling mediated by RhoA and Ca2+ is demonstrated as a key host-oriented mechanism for viral entry upstream of the signalling pathway. We now tackle the molecular identification of the interface between IAVs and host cells by using fluorescence bioimaging and the microchamber technology, in order to quantitatively analyse the event during virus infection.

3SHA-02 *in vitro* 系で明らかになった細菌べん毛形成の分子機構とその制御

Molecular mechanism of the flagellar biogenesis revealed by *in vitro* transport assay system

Katsumi Imada (*Grad. Sch. Sci., Osaka Univ.*)

The bacterial flagellar biogenesis is a well-controlled process regulated by various proteins, including a small number of multifunctional chaperons. The flagellum is an exracellular organelle, and most of the components are secreted through the flagellar type III secretion apparatus. Recent studies have revealed that the flagellar gene expression is closely coupled with the protein secretion. However, the molecular mechanism of the flagellar biogenesis is still obscure because the complicated regulatory feedbacks greatly affect the flagellar construction *in vivo*. We have developed an *in vitro* transport assay system using an inverted membrane vesicle (IMV). We will show and discuss the novel findings on the flagellar biogenesis obtained by the IMV assay.

3SHA-03 真核細胞の走化性における濃度勾配センシングと方向性のある細胞運動

Gradient sensing and directed cell migration in eukaryotic chemotaxis

Masahiro Ueda[1,2] ([1]*Graduate School of Sciences, Osaka University*, [2]*QBiC, RIKEN*)

We have studied chemotaxis of *Dictyostelium* cells as a typical example of the stochastic signal transduction. The cells can exhibit extreme sensitivity to chemical gradients over a wide range of concentrations, although the underlying mechanisms are only partially understood. Recently, we found that a novel regulator of heterotrimeric G proteins is essential for extending the chemotactic range. Our findings illustrate chemoattractant receptor regulates G protein redistributions between membrane and cytoplasm through the new regulator, which likely serves as a wide range sensing mechanism. We will discuss the possible mechanism by which cells can sense and transduce chemotactic signals under the strong influence of molecular noise.

3SHA-04 勾配感知におけるノイジーなシグナルの時間微分

Temoporal Differentiation of Noisy Signal in Gradient Sensing

Tetsuya Kobayashi[1], Ryo Yokota[1,2] ([1]*Institute of Industrial Science, University of Tokyo*, [2]*Research and Education Platform for Dynamic Living States*)

Gradient sensing is a process to obtain the information of spatial derivative of the chemical gradient in environment. While the derivative is computed within a cell by comparing the small spatial difference of the receptor activities on different locations of the membrane (known as spatial sensing), cells can also compute the derivative from temporal change in the activity induced by active movement within the gradient (know as temporal sensing).
Computing derivative with the temporal change in the signal is more non-trivial especially when the signal contains stochasticity due to the noisy receptor activities.
In this work, we use the information-theoretic approach to investigate the optimal strategy for noisy temporal sensing and its biological relevance.

3SHA-05 マイノリティージェノタイプ・細胞数分布を１細胞レベルで同定・定量する新技術

A new method for identification of minor genotype and measurement of cell-number distribution at the single cell level

Katsuyuki Shiroguchi (*IMS RIKEN*)

The small number of cells (minority) which have particular genotype, such as cancer cell, B cell recognizing an antigen, or virus, may affect cell populations, tissues, or individuals. The variety of minorities and their balance in number may also affect a biological system; for example, there are many bacteria in the intestine, called gut flora, which affect their host's state (health, etc). In order to investigate the significance of these minorities and their balance for biological systems, one of the first questions may be: how many of what type of cells exists? I will present our newly developed automatic cell barcoding method which enables one to identify genotype of many single cells in a high throughput manner within a next generation sequencer workflow.

3SHA-06 個体レベルの「時間」の理解に向けて ―全身・全脳透明化の先に見えるもの―
Towards System-level Understanding of Biological Time

Hiroki R. Ueda[1,2] ([1]*The University of Tokyo*, [2]*RIKEN, QBiC*)

Mammalian circadian clock system is a complex and dynamic system consisting of complicatedly integrated regulatory loops and displaying the various dynamic behaviors including i) endogenous oscillation with about 24-hour period, ii) entrainment to the external environmental changes (temperature and light cycle), and iii) temperature compensation over the wide range of temperature. I will discuss the current and past studies on a mammalian circadian clock as an example of molecule-to-cell-level systems biology, and also discuss the challenges and opportunities towards the organism-level systems biology. Especially, I will introduce the current update on the whole-brain and whole-body imaging with single-cell resolution as well as its biological applications.

3SIA-01 タンパク質の分子間相互作用への溶媒効果
Solvation effects on protein interaction with other molecules

Akio Kitao (*IMCB, Univ. Tokyo*)

Protein interaction with other molecules is a key to understand biological phenomena. Specificity and non-specificity of protein interactions depend on the structures of the interacting molecules and direct interactions as well as solvation and desolvation of the molecules. Therefore, investigation of solvation effects is essential to understand the mechanisms of protein complex formation. Computationally, it is not straightforward to directly evaluate solvation effects on large protein complexes such as protein-protein complex. We have been recently developing a method to evaluate protein complex models using all-atom molecular dynamics simulation combined with the solution theory in the energy representation, which will be reported in the symposium.

3SIA-02 Time-resolved resonance Raman observation of proteins in action

Yasuhisa Mizutani (*Osaka Univerisity*)

Resonance Raman (RR) spectroscopy is a versatile spectroscopic technique for studying the structure and dynamics of proteins. For chromoproteins, Raman bands of the chromophore are selectively enhanced when the excitation wavelength in visible wavelength region is employed. On the other hand, when the excitation wavelength is tuned to ultraviolet wavelength region, strong RR scattering from the peptide backbone and aromatic amino acids provides vibrational information on protein structure and environmental changes. Thus, we can selectively obtain structural information for the chromophore moiety and protein by tuning the excitation wavelength for RR measurements. In this talk, I will present our work on structural dynamics of light-driven ion pumps in their photocycles.

3SIA-03 タンパク質構造に対する環境効果のエネルギー相関解析
Correlation Analysis of Environmental Effect on Protein Structure with Explicit Solvent

Nobuyuki Matubayasi (*Division of Chemical Engineering, Graduate School of Engineering Science, Osaka University*)

The effect of temperature elevation on protein structure is analyzed from the standpoint of energetics using MD simulation combined with a theory of solvation. Through all-atom analysis of the solvation free energy over a wide range of protein structures, the relative stabilities of folded and unfolded structures are examined in terms of the correlations against the electrostatic, van der Waals, and excluded-volume components in the intramolecular and intermolecular interactions of the protein. It is found that the preference of the protein structure is in correspondence to the van der Waals and excluded-volume components in a large-scale variation of the structure. The effect of cosolvent is also addressed in connection to the urea denaturation.

3SIA-04 極限環境生物がデザインした蛋白質
Proteins designed by extremophiles

Yoshihiro Sambongi (*Hiroshima University*)

Extremophiles are defined as organisms that can survive environments hostile to mesophiles, or organisms which grow only in intermediate environments [Macelroy, Biosystems 6, 74-74 (1974)]. Proteins of extremophiles are designed so as to survive under such hostile conditions. For example, cytochrome c from a thermophile has a unique structure that is not found in its mesophilic counterpart. In addition, a halophilic pyrophosphatase that contains more acidic amino acid residues than its non-halophilic homologous enzyme functions only under the high salt concentrations. Comparative studies on the designs of homologous proteins from extremophiles and non-extremophiles have enhanced a greater understanding of protein structure and function.

3SIA-05 ヘリカルリピートタンパク質の細胞内での構造と機能
Intracellular structure and function of helical repeat proteins

Shigehiro Yoshimura, Hide Konishi, Suguru Asai (*Grad. Sch. Biostudies, Kyoto Univ.*)

Helical repeat proteins are composed of tandem arrays of α-helices and account for more than 5 % of annotated proteins in humans. They are classified into several distinct motifs such as ARM repeat, Tetratricopeptide repeat (TPR), HEAT repeat, and pumilio homology domain. Although these motifs are structurally similar to each other and mostly participate in protein-protein and protein-nucleic acid interactions, they are involved in a large variety of cellular processes, such as protein transport, chromosome condensation and signal transduction. In this study, we investigated the structural properties of these proteins in various environments including aqueous solutions and macromolecular crowding, and revealed their unique structural stability and flexibility.

3SIA-06 バクテリア細胞質中の蛋白質および代謝物のダイナミクス・安定性・相互作用：全原子分子動力学法による理論的研究
Dynamics, Stability, and Interactions of Proteins and Metabolites in Bacterial Cytoplasm: All-atom Molecular Dynamics Study

Isseki Yu[1,4], Takaharu Mori[1], Tadashi Ando[2], Ryuhei Harada[3], Jaewoon Jung[3], Yuji Sugita[1,2,3,4], Michael Feig[5] ([1]*RIKEN Theoretical Molecular Science Laboratory*, [2]*RIKEN QBiC*, [3]*RIKEN AICS*, [4]*RIKEN iTHES*, [5]*Department of Biochemistry and Molecular Biology, Michigan State University*)

How biomolecules are working in the cellular environment is one of the most fundamental questions in life science. We constructed full atomistic model of the cytoplasm of bacteria (Mycoplasma genitalium) covering 10% of an entire cell. Using this model, we performed molecular dynamics (MD) simulation with the highly parallelized MD program GENESIS on K computer. The simulations provide information of protein dynamics, stability, and interactions in cell. The protein diffusion was consistent with the experimental data. Metabolite dynamics are also investigated and it was found that most of ATPs show two-dimensional diffusion on the protein surface. The current simulation opens a new era to connect our understanding between molecular and cellular levels in biology.

3SIA-07 NMR を使ったジスルフィド結合の細胞内解析
Stability of disulfide bonds of proteins in the cytosolic space analyzed using NMR spectroscopy

Shuhei Murayama[2], Yoshiaki Enokizono[1], Ken-ichi Akagi[3], Naotaka Sekiyama[1], Kohsuke Inomata[4], Masahiro Shirakawa[2], **Hidehito Tochio**[1] ([1]*Graduate School of Science, Kyoto University*, [2]*Graduate School of Engineering, Kyoto University*, [3]*National Institute of Biomedical Innovation*, [4]*RIKEN*)

Disulfide(SS) bonds are critical in maintaining 3D structure and function of proteins, the stability of which depends on the redox potential of the surrounding environment. In general, as the cytosol is in a reductive condition, solvent exposed SS bonds are readily reduced to the SH form. However, in some cases SS bonds are not exposed but well integrated in the structure, making them relatively stable. Those SS bonds would be partly maintained even in the cytosol, providing an opportunity for functional switching of the protein, because cytosolic redox potential depends on the cellular state and oxidative stress. To evaluate the stability of SS bonds in cells, which will also rely on molecular crowding effect, in-cell NMR analyses were performed on model proteins.

3SJA-01 ストリング法を用いた多剤排出トランスポーター AcrB の薬剤排出機構の解析
Drug extrusion mechanism of multidrug exporter AcrB studied by the string method

Yasuhiro Matsunaga (*RIKEN AICS*)

The multidrug transporter AcrB actively extrudes a wide spectrum of noxious compounds out of the bacterium using proton-motive force. The AcrB is an asymmetric homotrimer whose monomers undergo cyclic conformational changes during drug export; this process is referred as functional rotation. We investigated the mechanism of the functional rotation by using atomistic molecular dynamics simulations. A single step of functional rotation was simulated with the string method, and its free energy landscape was evaluated. Comparison of different protonation states suggested that protonation in the transmembrane region induces a conformational change of the porter domain. Atomic details of this conformational change and drug extrusion process will be reported.

3SJA-02 大規模分子集合体系におけるレアイベントの分子動力学計算
Molecular dynamics study of rare events of large-scale molecular systems

Noriyuki Yoshii (*Grad. Sch. Eng., Nagoya Univ.*)

Nowadays the use of both peta-FLOPS class massively parallel supercomputers and general purpose software available for large scale computation allowed large-scale molecular dynamics (MD) calculation of more than 10 million atoms. This enables us to perform μ second order long-time MD calculation of large biomolecular systems such as virus and lipid vesicles. However, in general, time constant of biological function is extremely longer than that of MD calculation. It is quite difficult to obtain dynamic aspects of biomolecules from simple MD calculations. In this presentation I will talk our recent result about the empty virus capsid as an example of the large-scale MD calculation. I will also mention about our study to estimate time constant of the rare event.

3SJA-03 緩和モード解析による蛋白質の動的性質の研究
Exploring Dynamics and Kinetics of Proteins using Relaxation Mode Analysis

Ayori Mitsutake[1,2] ([1]*Dep. of Phys., Keio Univ.*, [2]*JST, Presto*)

Classical molecular dynamics (MD) simulation is a popular and powerful method for describing the structure, dynamics, and function of proteins at atomic resolution. Recent technological advances have allowed us to perform long simulations. As longer and larger MD simulations are performed, it is more important to develop analysis methods to investigate dynamics or kinetics of proteins. Relaxation mode analysis (RMA) were developed to investigate “dynamic” properties of polymer, homo-polymer, systems. In RMA, slow relaxation modes are extracted from MD simulations. Recently, RMA has been applied to proteins, hetero-polymer systems to investigate dynamic properties of structural fluctuations. Here, we explain RMA and show the results of some proteins studied by RMA.

3SJA-04 生物学的レアイベントを再現する効率的構造サンプリング手法
Simple yet powerful conformational sampling methods for reproducing biologically rare events

Ryuhei Harada (*CCS, Univ. of Tsukuba*)

Biologically rare events are related to functions of proteins. In the most of biological processes, the rare events are observed as conformational transitions related to the biological functions. In this study, we propose several powerful conformational sampling methods by repeating of (I) Selections of seeds and (II) Conformational resampling from the seeds. Our methods are referred to as PaCS-MD, FFM, TBSA, and OFLOOD, respectively. In each method, measures are specified to identify the seeds that have high potential to transit, and followed by the conformational resampling by restating short-time MD simulations from the seeds. In this presentation, we show our methods drastically promote conformational transitions, reproducing the rare events efficiently.

3SJA-05 時間分解共鳴ラマン分光法をもちいたレチナールタンパク質におけるレアイベント観測

Observation of rare events in retinal proteins revealed by time-resolved resonance Raman spectroscopy

Misao Mizuno (*Graduate School of Science, Osaka University*)

Protein functions are regulated by rare events such as structural changes. Knowledge of protein motions is important for determining mechanism of protein functions. For retinal proteins, light absorption induces local structural changes in a retinal chromophore as a functional trigger. The local change produces sequential changes in the higher order structure, thereby facilitating function. To elucidate the functionally important rare events in retinal proteins, we observed structural dynamics by using time-resolved resonance Raman spectroscopy. We explored picosecond protein responses to the photoisomerization of the chromophore as well as structural evolutions of the retinal chromophore from nanoseconds to milliseconds.

3SJA-06 天然変性領域にレアに生じる構造が持つ役割

Role of rarely happening fold in intrinsically disordered proteins (IDPs)

Shin-ichi Tate[1,2] ([1]*Dept. Math. and Life Sci., Hiroshima Univ.*, [2]*RcMcD, Hiroshima Univ.*)

Intrinsically disordered proteins (IDPs) or regions (IDRs) have been recognized to play essential roles that the structural parts of proteins cannot achieve. IDPs sample a wide range of conformations, in which process they transiently stay in folded or locally structured states. The folded states are not stable, but rapidly back to the other unstructured states. The populations of the folded structures are, therefore, essentially low. In this sense, the transient folding of IDP is 'rare event'.
In this talk, I will describe how the transiently folded structures in the IDPs role in functional regulation, using the examples from the studies on the IDPs currently running in my laboratory.

3SKA-01 人工細胞構築のための細胞サイズリポソームの生成と応用

Preparation and application of cell-sized liposomes for synthesis of artificial cells

Masamune Morita[1,2] ([1]*Interdisciplinary Grad. Sch. Sci. and Eng., Tokyo Tech.*, [2]*JSPS Research Fellow*)

The synthetic approach to biological systems is an important and interesting challenge in biophysics and biochemistry. Cell-sized liposomes, as artificial cell membrane systems, are a powerful tool for clarifying physicochemical mechanisms of structural membrane dynamics. To efficiently use them, it is required more complex membrane structure, such as microdomain formation or asymmetry of inner/outer leaflets in a lipid-bilayer membrane. In this presentation, I show the simple production method of cell-sized liposomes with complex membrane structure by centrifugal microfluidic device. Moreover, I show the membrane structural dynamics of artificial cell membrane by external molecules. Finally, I would like to discuss the outlook of the future in artificial cells.

3SKA-02 ボトムアップアプローチによる人工細胞の作製

Preparation of artificial cell models by bottom-up approach

Koki Kamiya[1,2] ([1]*Kanagawa Academy of Science and Technology*, [2]*PRESTO, JST*)

The development of artificial cell models has been studied intensively. These studies play a role in the understanding of the origin of life and the molecular mechanisms underlying biological reactions in cells. Giant lipid vesicles and giant liposomes are suitable materials that can be used to build these cell models. However, the constrictions of giant vesicles, which have complex cellular functions such as signal transduction, have been difficult: there is a lack of reconstitution methodology for controlling the lipid components and the orientation of membrane proteins. In this presentation, I will introduce a method for reconstituting membrane proteins, which maintains the original orientation of these proteins.

3SKA-03 細胞サイズ閉鎖空間内でのアクチン細胞骨格の再構成

Reconstitution of actin cytoskeleton in a cell-sized confined space

Makito Miyazaki[1], Masataka Chiba[1], Hiroki Eguchi[1], Takashi Ohki[1], Shin'ichi Ishiwata[1,2] ([1]*Dept. of Physics, Waseda Univ.*, [2]*WABIOS, Waseda Univ.*)

Animal cells possess various kinds of cytoskeletal structures composed of actin filaments and myosin motors including actomyosin cortex and cytokinetic rings. Although the accessory proteins that regulate the formation of actin cytoskeleton have been identified step by step, the self-assembly mechanism of actin cytoskeleton remains unclear. Here, we developed simple *in vitro* model systems by encapsulating purified actomyosin into cell-sized water-in-oil droplets or liposomes. By controlling the protein components and concentrations, ordered patterns reminiscent of actomyosin cortex and cytokinetic rings were self-organized. We will present the preparation method and the dynamics of *in vitro* model systems, and discuss the assembly mechanisms of actin cytoskeleton.

3SKA-04 ミクロ水滴から出発する自己駆動型人工細胞へのアプローチ

Approach to self-propelled artificial cells from microdroplets

Hiroaki Ito, Masatoshi Ichikawa (*Grad. Sch. Sci., Kyoto Univ.*)

Active force generation in living organisms plays crucial roles in various biological processes, such as cell division and cell motility. In particular, the implementation of the dynamic interfacial deformability driven by this active force remains as an essential step to reconstitute self-propelled artificial-cell systems. In this talk, we (i) demonstrate how the force generated from cytoskeletal and motor proteins can contribute to dynamic deformation of a lipid monolayer coating a microdroplet, and (ii) propose a strategy to develop the lipid monolayer system into a lipid bilayer system. To understand the underlying mechanisms of the experimentally observed phenomena will shed light on how biological macromolecules can contribute to the artificial-cell systems.

3SKA-05 人工細胞を活用した高分子ミクロゲル形状の物理的な制御
Shape control of biopolymer microgels utilizing model cells

Miho Yanagisawa (*Dept. Appl. Phys.,Tokyo Univ. of Agri. & Tech.*)

Phase separation and gelation of biopolymers plays important roles in regulating cellular structure. We mimic such structural formation of biopolymers using model cells, and establish a way of microgels molding upon phase separation, wetting, and gelation of the polymers. We confined a polymer blend of gelation polymer and non-gelation polymer in microdroplets coated with a lipid layer. Temperature shift to trigger gelation and phase separation produces two types of microgels, i.e., microcapsules and hemisphere microgels according to complete and partial wetting of the polymer. In addition, the wetting of the gelation polymer affects elasticity of the microgels. This method to regulate shape and elasticity of microgels will broaden fields of the microgel application.

3SKA-06 膜タンパク質を人工細胞に組み込み、進化させる
Installation and directed evolution of membrane proteins in artificial cells

Satoshi Fujii (*Grad. Info. Sci., Osaka Univ.*)

Reconstitution of artificial cells by encapsulating the cell-free translation system and genes of interest in liposomes had generated many life-like reactions in test-tubes. For further development, we have reconstituted the “responsiveness” by using alpha-hemolysin, a membrane protein derived from Staphylococcus aureus. So far, we found a membrane curvature dependency of alpha-hemolysin, which implied the importance of the artificial cell size or shape for membrane protein studies. Moreover, we developed a method, “liposome display”, which enables the directed evolution of membrane protein in vitro, and succeeded to generate a mutant of alpha-hemolysin with higher activity.

3SKA-07 細胞再構成へ：生細胞に近い人工細胞の創成
Creating Life-mimicking Artificial Cells toward re-building living cells

Kei Fujiwara (*Dept. Biosciences and Informatics, Keio University*)

Reconstitution of living cells from “non-alive” biomolecules is a big challenge in life science. We have been addressing this challenge by creating artificial cells and by analyzing them to reveal the critical features of life. So far, we have developed a method to prepare an additive-free cell extract, and to concentrate the macromolecules inside liposomes by using osmotic pressure. This method enabled us to create a life-mimicking artificial cell (L-MAC), which are liposomes entrapping the highly-concentrated biomolecules reaching physiological concentration. The most remarkable feature of L-MACs is quite slow diffusion of biomolecules. We introduce the method to prepare L-MACs, and discuss the upcoming research of cell reconstitution beyond the recent results.

3SKA-08 合成細胞生物学ツールとして利用可能な GUV 人工細胞モデル
GUV-based artificial cell model available as constructive cell biology tool

Shin-ichiro Nomura (*Department of Bioeng. Robotics., Tohoku Univ.*)

Recently, we have become to call a complex structure of lipid vesicle as an artificial cell which incorporating live-cell-like function(s). Here we discuss about our two examples available as a research tool. Improving vesicle preparation method enables to encapsulate various hydrophilic materials inside of giant unilamellar vesicle (GUV). We showed that electrofusion between GUV and live cell enables transferring GUV contents (~um sized) into cellular cytosol. We are also trying to construct an artificial molecular robot of 2nd generation. Designed molecular devices of actuator (motor proteins) and controller (DNA circuits) are installed into GUV. These examples of both cell-alternation and bottom-up construction will contribute for constructive-cell biology.

1C1320 アミロイドβ1-42多量体の構造ダイナミクスの高速AFM観察

Video imaging of structural dynamics of individual amyloid β 1-42 aggregates

Masahiro Itami[1], Kenjiro Ono[2], Takahiro Nakayama[3] ([1]*Grad. Sch. Sci., Univ. Kanazawa*, [2]*Kanazawa Univ. Hospital*, [3]*Kanazawa Univ. Bio AFM FRC*)

Amyloid fibril formation is related to various conformational disease. Now in the fibril growth, nucleation-dependent and dock-lock monomer incorporation models are generally accepted. However, there was nothing to directly observe growing fibril structure in real time. To reveal structural change in these models, we tried to observe aggregation and dissociation of Amyloid β (Aβ) 1-42 involved in Alzheimer's disease (AD). We succeeded to observe changing of fibril structure and dissociation of large aggregates. Our results provide the insights into the dock-lock mechanism.

1C1335 ユビキチンリガーゼ (HECT型 E3) のユビキチン化に伴う動態の高速AFM観察

Observation of the dynamics associated with ubiquitination of HECT E3 ubiquitin ligase using High speed AFM

Fuminori Kobayashi[1], Takamitsu Haruyama[2], Takahiro Nakayama[2], Noriyuki Kodera[2], Hiroki Konno[2] ([1]*Grad. Sch. Sci., Univ. Kanazawa*, [2]*Kanazawa Univ. Bio AFM FRC*)

Ubiquitin conjugation to the target protein (Ubiquitination) is one of the post-translational modifications. Ubiquitination is accomplished through catalytic cascade by E1, E2 and E3. HECT-type E3 has the HECT domain that forms intermediate thioester bind with ubiquitin before catalysing the substrate ubiquitination. HECT domain is composed of N- and C-lobe and flexible hinge loop connecting these lobes. It has been suggested that flexibility of the hinge loop enable conformational change in HECT domain and is therefore required for ubiquitin transfer. To investigate the effect of the flexibility of hinge loop for ubiquitin transfer, we directly visualized the movement of C-lobe both in wild type and hinge loop mutant using High-speed AFM.

1C1350 高速AFMによる抗体のやわらかさ測定

High-Speed AFM reveals swinging nature of antibody with flexible arms

Norito Kotani, Kumaresan Ramanujam, Yoko Kwamoto, Takao Okada (*Research Institute of Biomolecule Metrology Co.,Ltd.*)

High-speed Atomic Force Microscopy (HS-AFM) has been applied to observe dynamic behavior of biomolecules as movie, without any special treatments. We have observed the dynamic behavior of IgG in solution using HS-AFM. "Y" shape of IgG was imaged clearly, and the Fab and Fc regions were precisely distinguished. The Fab regions moved in torsional direction like swinging arms as we observed in HS-AFM movie. Flexible structure of hinge regions is responsible for this dynamic behaviour. This flexibile nature contributes for the easy binding of IgG to the antigen. For the first time, we have identified the swinging nature of this soft structure, which is important in antibody functioning.

1C1405 高速原子間力顕微鏡の温度制御機構の開発と好熱菌FliIの観察

Development of Temperature Controlled High-Speed AFM and Observation of Thermus Thermophilus FliI

Kei Adachi[1], Takayuki Uchihashi[1], Katsumi Imada[2], Ken Yokoyama[3], Toshio Ando[1] ([1]*Coll. Sci. & Eng., Kanazawa Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Facul. Biosci., Kyoto Sangyo Univ.*)

High-speed atomic force microscopy (HS-AFM) is a powerful tool for biomolecular studies as it can capture high resolution images of single biomolecules in dynamic action. However, the operation temperature of the current HS-AFM is limited to room temperature even though higher temperature is optimum for the physiological activity of some proteins, in particular those from thermophiles. To extend the application range of HS-AFM, we developed temperature-controlled HS-AFM and confirmed that this HS-AFM can be operated over 40 °C without disturbance of the imaging. Using this system, we observed conformational dynamics of hexameric ATPase FliI derived from Thermus Thermophilus that has an optimum temperature around 40 °C for the ATPase activity.

1C1425 ヘテロな系でのAFMの応用に向けたAFM・TEMの相関顕微鏡法

Correlative Atomic Force and Electron Microscopy toward Applications of Atomic Force Microscopy to Heterogeneous Systems

Yutaro Yamada[1], Takamitsu Haruyama[1], Hiroki Konno[1], Katsuya Shimabukuro[1] ([1]*UNCT*, [2]*Bio-AFM, Kanazawa Univ.*)

AFM is one of the most powerful systems to investigate specimens at nano-meter temporal resolution. Applications of AFM, however, have been limited to simple systems due to the lack of the ability to discern molecules. Many essential biological phenomenon a set of proteins to function properly. Therefore, technologies to analyze such heterogeneous systems are highly demanded. Here, we propose a new method called nanoCAFE (nano correlative atomic force and electron microscopy) to extend the AFM function to heterogeneous systems by combining AFM and electron microscopy. Here, by merging AFM and EM we are able to investigate molecules in two different microscopies. Thus, nanoCAFE could be a powerful method to dissect complicated biological phenomenon.

1C1440 大球コロイドの周りに分布している小球コロイドの数密度分布の計測理論

Measurement theory of density distribution of small colloids around a large colloid

Ken-ichi Amano, Kota Hashimoto, Naoya Nishi, Tetsuo Sakka (*Graduate School of Engineering, Kyoto University*)

Recently, we proposed a theory that transforms a force curve between two large colloids into the density distribution of small colloids around the large colloid. The force curve is obtained by using laser tweezers, colloidal probe atomic force microscopy, light scattering, etc. We conducted a verification test of the theory in a computer, and found that the theory can reproduce the density distribution precisely when the volume fraction of the small colloids is low. However, when the volume fraction is high, the reproducibility is not so high. In the presentation, we explain the theory in detail and show the results of the verification test. Applicability of the theory to the experiment is also discussed.

1C1455 ナノスケールの形状・化学物質濃度プロファイルを可視化するナノ電気化学顕微鏡の創成

若手招待講演

Development of Nano Electrochemical Microscopy for Visualizing Nanoscale Cell Surface Topography and Chemical Profile

Yasufumi Takahashi[1,2,3], Hiroki Ida[2], Hitoshi Shiku[2], Tomokazu Matsue[1,2] ([1]*WPI-AIMR of Tohoku University*, [2]*Environmental studies, Graduate school of Tohoku University*, [3]*JST PREST*)

To link the function and structure of cell, chemical concentration profile is important. We developed high resolution scanning electrochemical microscopy (SECM) for mapping the chemical concentration profile around the cell surface. SECM uses microelectrode as a probe and image the chemical concentration profile from a redox current. The electrode-sample distance control is important for accurate current measurement because the current signal is influenced by the surface roughness.
We developed ion current as a feedback SECM (NanoSECM) to visualize topography and chemical concentration profile simultaneously. We measured neurotransmitter release, variation of membrane expression level, and progress of differentiation process of ES cells.

1C1510 Spectral fingerprinting of individual cells observed by cavity-reflection-enhanced light-absorption microscopy

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The absorption spectrum is known as a “molecular fingerprint”, which would be useful to investigate the cellular status. However, cells are too thin for their absorption measurement. Here, we developed an optical-cavity-enhanced absorption spectroscopic microscopy. The light absorption is enhanced by an optical cavity system, which allows the detection of the absorption spectrum with samples having an optical path length of 10 μm at sub-cellular spatial resolution. Principal component analysis suggests the cellular individuality. Furthermore, this microscopy allows to observe frozen sections of tissue samples without staining. Thus, our microscopy opens the door for imaging the absorption spectra of biological samples and thereby detecting the individuality of cells.

1C1530 細胞観察に向けたティップスキャン型高速 AFM の改良

Improvement of tip-scan HS-AFM for live-cell imaging

Shin-nosuke Yamanaka[1], Hiroki Watanabe[1,2], Takayuki Uchihashi[1,3], Toshio Ando[1,3] ([1]*Grad. Sch. Sci, Univ. Kanazawa*, [2]*RIBM*, [3]*Bio-AFM Center*)

Recently we have developed the combined system between high-speed AFM (HS-AFM) and fluorescence microscopy with which conformational dynamics of single protein and fluorescent spot can be simultaneously observed [1]. However, in this system the observation area is limited to less than 5 μm and therefore hard to be applied to large biological samples such as live cells. This limitation is due to the limited operation range of the laser tracking system. We improved the laser tracking system to extend the scanning range to several tens of micrometers. In the presentation, we will demonstrate the performance of the extended combined system and live-cell imaging with this system.
References
[1] S. Fukuda et al., Review of Scientific Instruments 84, 073706 (2013)

1C1545 Long-tip 高速原子間力顕微鏡による生きた細胞の形態観察

Live-cell imaging by long-tip high-speed atomic force microscopy

Mikihiro Shibata[1,2], Takayuki Uchihashi[2,3], Toshio Ando[2,3], Ryohei Yasuda[1] ([1]*MPFI*, [2]*Dept. Phys., Kanazawa Univ.*, [3]*Bio-AFM, Kanazawa Univ.*)

High-speed atomic force microscopy (HS-AFM) is capable of direct visualization of conformational changes of single proteins under physiological conditions. However, the application for imaging of live mammalian cells has been complicated because of the collision between the cantilever and cells. To apply HS-AFM to live mammalian cells, we have developed an extremely long (~3 μm) and thin (~5 nm) AFM tip to avoid the collision. Also, we have combined HS-AFM with fluorescence microscopy to locate the AFM tip on the region of interest. After these optimizations, we suceeded in imaging the morphogenesis of filopodia, membrane ruffles, pit formation, and endocytosis in COS-7 and HeLa cells, as well as dissociated hippocampal neurons.
M. Shibata et al. (2015) *Sci. Rep.*

1C1600 動物細胞の 1 細胞系譜の取得に向けたマイクロ流体デバイス

Microfluidic device for tracking mammalian cells along single-cell lineages

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In general, the phenotypes of individual cells in clonal population are heterogeneous and fluctuate in time. Obtaining the information of the unique cell lineages that, for examples, differentiate in response to external signals or that adapt to severe stress such as antibiotic exposure requires tracking single-cell lineages in population across changing environments. Here, we developed a new microfluidic device for long-term tracking of mammalian cells and observed lymphocytic cells (L1210) over 35 generations in a constant environment. The result revealed that cells stably grow in this device with the mean generation time 14 hours, which is consistent with the batch culture data. We are now investigating the response of L1210 cells to anticancer drug by this device.

1D1320 単量体・二量体平衡の定量的解析による Photozipper の分子機構解明

Quantitative analyses of the monomer-dimer equilibrium reveal the molecular mechanism of Photozipper

Yoichi Nakatani, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

Photozipper (PZ) is a blue light-regulated dimerizing module consisting of a basic leucine zipper (bZIP) and a light-oxygen-voltage sensing (LOV) domains. In this study, four different PZ constructs were prepared comprising different N-terminal truncations, and the monomer-dimer equilibria of PZs were investigated in the dark and light states. Dynamic light scattering and size exclusion chromatography analyses revealed that ZIP region stabilized the monomeric form in the dark state. In the light state, FRET analyses demonstrated that intermolecular LOV-LOV and ZIP-ZIP interactions stabilized the dimeric forms. Our results suggest that synergistic interactions between the LOV and bZIP domains possibly confers to the function of PZ as a BL-regulated molecular switch.

1D1335 (6–4)光回復酵素の拡張された電子移動経路

An expanded electron transfer pathway in the (6-4) photolyase

Junpei Yamamoto[1], Pavel Müller[2], Kohei Shimizu[1], Klaus Brettel[2], Shigenori Iwai[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*CEA Saclay, France*)

Photolyases (PLs) are flavoproteins that directly repair cyclobutane pyrimidine dimers (CPDs) and pyrimidine(6-4)pyrimidone photoproducts ((6-4)PPs) by utilizing the blue light. Among them, the (6-4) photolyases are well-related to cryptochromes, which are involved in regulation of circadian clock, and they share high homologies in amino acid sequences, chromophore, and tertiary structures, although their functions are diverse. In this study, we focused on an amino acid residue conversed among vertebrate/insect (6-4) photolyases and vertebrate cryptochromes and discovered its involvement in the electron transfer pathway in the photoactivation and photorepair.

1D1350 CPD 光回復酵素と(6-4)光回復酵素の機能転換

Functional conversion of CPD and (6-4) photolyases by mutation

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Photolyases (PHRs) are DNA repair enzymes. Two types of PHRs have been reported: CPD-PHR repairs cyclobutane pyrimidine dimers (CPDs), while (6-4)PHR repairs (6-4) photoproducts. Their sequence identity is ~25 %, which provides specific function to repair each substrate. In this study, we attempted functional conversion between CPD and (6-4) PHRs, whose repair signals were monitored by FTIR spectroscopy. We found that a triple mutant of (6-4) PHR can repair the CPD photoproduct. In contrast, the (6-4) photoproduct was not repaired by the reverse triple mutation of CPD PHR, even after 8 more mutations were added. Molecular mechanisms of asymmetric functional conversion will be discussed based on the present experimental and computational results.

1D1405 シロイヌナズナクリプトクロム 1 の光反応における赤外分光研究

FTIR study of the Arabidopsis Cryptochrome1 photoreaction

Katsuhiro Mikuni[1], Daichi Yamada[1], Tatsuya Iwata[1], Kenichi Hitomi[2], Elizabeth D. Getzoff[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*The Scripps Res. Inst. USA*)

Cryptochrome is a blue-light receptor for photomorphogenesis in plants. It is known that cryptochrome 1 from Arabidopsis (AtCRY1) can bind ATP, which promotes a photoactivation (reduction of FAD) in vitro. In order to investigate how ATP influences structural changes in AtCRY1 during the photoactivation, we compared light-induced difference FTIR spectra of AtCRY1 in the absence and presence of ATP. Differences were observed in the amide I region, which reflects the secondary structure of peptide backbone. We also observed deprotonation of a carboxylic acid only in the presence of ATP. Molecular mechanism of CRY activation and role of ATP will be discussed.

1D1425 視物質の低い熱活性化頻度をもたらす分子メカニズム

Molecular mechanism of the low thermal activation rate of visual pigments

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Most vertebrates have two types of photoreceptor cells, rods and cones, which are responsible for scotopic and photopic vision, respectively. Rods show a low threshold of photon detection. Therefore, the low thermal activation rate (kth) of rhodopsin, rod visual pigment, is crucial for the function of rods. Phylogenetic analysis indicates that rhodopsin has evolved out of cone visual pigment, but the molecular mechanism of the low kth of rhodopsin is unknown. We determined key amino acid residues which control the kth of rhodopsin and frog blue-sensitive cone visual pigment, which is expressed in rods and shows the low kth, by biochemical and spectroscopic measurements. Based on our results, we discuss the common mechanism of the low kth of visual pigments.

1D1440 レチナール異性化によるロドプシン活性化のメカニズムの解析

Mechanism of how retinal isomerization changes the structure of rhodopsin to the active state

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Rhodopsin consists of a protein component opsin and an 11-*cis* retinal chromophore. We have studied the mechanism for how retinal *cis-trans* isomerization triggers rhodopsin activation. NMR distance measurements between specific carbons of the retinal and the amino acids surrounding it provide information on the orientation of the all-*trans* retinal in the active state. NMR chemical shift changes and FTIR measurements show changes in hydrogen bonding interactions involving residues in EL2. We propose that Y191 shifts toward E181 upon activation and decreases the contact with TM6, which is not shown in the crystal structure of active opsin. This change of the EL2 and TM6 interaction facilitates the outward rotation of TM6, which is a common movement among class A GPCRs.

1D1455 青感受性視物質における Y265 の役割

Role of Y265 in blue-sensitive visual pigment

Yuki Nonaka[1], Kota Katayama[1,2], Kei Tsutsui[3], Hiroo Imai[3], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Dept. Pharm., CWRU, USA*, [3]*Primate Res. Inst., Kyoto Univ.*)

Primates including human have three types of color visual pigments; blue, green, and red. Although these pigments contain an identical chromophore molecule, 11-cis-retinal, different chromophore-protein interactions allow absorption of different colors. A key residue for spectral blue shift is Tyr265 in blue-sensitive visual pigment, whose corresponding amino acid is Trp in green- and red-sensitive visual pigment and rhodopsin.
In this study, we prepared Y265W mutant in monkey blue-sensitive visual pigment and W265Y mutant in monkey rhodopsin. The mutation effect was monitored by light-induced FTIR spectroscopy at 77K. We will discuss how blue-light absorption is determined structurally in blue-sensitive visual pigment.

1D1510 サル緑感受性視物質に対する陰イオン効果の構造研究
Anion effect to monkey green studied by light-induced difference FTIR spectroscopy

Shunta Nakamura[1], Kota Katayama[1,2], Hiroo Imai[3], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Dept. Pharm., CWRU, USA*, [3]*Primate Res. Inst., Kyoto Univ.*)

Primate green visual pigment is a G-protein-coupled receptor, which binds 11-cis retinal as a chromophore. The absorption maximum (λ_{max}) of monkey green (MG) is red-shifted dependent on chloride concentration. On the other hand, nitrate bound form exhibits an about 40 nm blue shift in the λ_{max}. In this study we compared light-induced difference FTIR spectra of chloride and nitrate bound MG at 77K. Several vibrational band difference for nitrate, protein and internal water, based on which local structure of the chloride-binding site of MG will be discussed.

1D1530 オプシン発現培養細胞の生化学的な応答に基づく非視覚型オプシンの分光感度の推定
Estimating spectral sensitivities of non-visual opsins based on biochemical responses of opsin-expressing cultured cells

Tomohiro Sugihara[1], Takashi Nagata[1], Mitsumasa Koyanagi[1,2,3], Akihisa Terakita[1,2] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*OCARINA*, [3]*JST-PRESTO*)

Spectral sensitivity of an opsin, a light-sensor protein in animals provides basic and important information to speculate its relevant physiology but spectral characteristics of not a few opsins still unknown. Basically, we express opsins in cultured cells and obtain functional recombinant opsin-based pigments. However, lower expression level of opsins often limits to analyze their absorption spectra. We have recently succeeded in quantitatively analyzing wavelength-dependent second messenger changes in cultured cells expressing opsins even in lower level. Here, by analyzing the second messenger changes, we determined spectral sensitivities of some non-visual opsins that are functionally important but had not yet been investigated for their absorption characteristics.

1D1545 TMT1 オプシンと TMT2 オプシンの分子特性比較解析
Comparative studies on the molecular properties between TMT1 and TMT2 opsins

Kazumi Sakai, Takahiro Yamashita, Yasushi Imamoto, Yoshinori Shichida (*Grad. Sch. Sci., Kyoto Univ.*)

Vertebrate Opn3/TMT opsins are photoreceptive proteins which are expressed in not only eyes and brains but also other tissues and phylogenetically divided into four subgroups (TMT1, TMT2, TMT3 and Opn3/Encephalopsin). We obtained recombinant proteins of TMT1 and TMT2 opsins and compared their molecular properties. These two TMT opsins were blue light sensitive Gi/Go-coupled receptors but had different active states. The active state of TMT1 opsin is a visible light-absorbing intermediate and can interconvert with the original and other resting states by light absorption, whereas that of TMT2 opsin is a UV light-absorbing intermediate and cannot convert back to the resting state. We will discuss the diversity of the activation mechanism in Opn3/TMT opsin group.

1D1600 G_s タンパク質の光制御に向けたキメラタンパク質の創出
Construction of chimeric proteins for optical control of G_s-protein activity

Kazuho Yoshida[1], Keiichi Inoue[1,2], Takahiro Yamashita[3], Rei Abe-Yoshizumi[1], Mizuna Tanaka[1], Kengo Sasaki[1], Yoshinori Shichida[3], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*JST PRESTO*, [3]*Grad. Sch. Sci., Univ. Kyoto*)

G-protein-coupled receptors (GPCRs) are heptahelical transmembrane receptors, which transduce signals through specific G-proteins to intracellular signaling cascades. We attempt to create new chimeric proteins using microbial rhodopsins for the optical control of G-protein signaling.
In this study, we designed chimeras between *Gloeobacter* rhodopsin, a proton pumping microbial rhodopsin, and β_2-adrenergic receptor for G_s-activation. These chimeras showed light-dependent G_s-activation by *in vitro* G-protein activation assay. We now try to measure intracellular G_s-activity using fluorescent imaging. We will discuss specific properties obtained from spectroscopic measurements and possible application to optogenetics.

1E1320 GTP 加水分解と PKC リン酸化によるダイナミン–コルタクチン複合体の制御
Regulation of Dynamin-Cortactin complex by GTP hydrolysis and PKC phosphorylation

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Dynamin-Cortactin complex is implicated in actin bundle formation required for variety of cellular events. Although the complex is thought to mechanically bundle actin filaments through the use of conformational change upon GTP hydrolysis by Dynamin, the precise mode of action of remains to be elucidated. To clarify mechanisms of complex assembly and conformational alteration of Dynamin-Cortactin complex, the proteins were incubated in a variety of nucleotide conditions that mimic reaction steps in GTP hydrolysis and examined by negative stain EM. High-speed AFM was also employed to visualize real-time dynamics. In addition, structural changes of the complex by PKC phosphorylation of cortactin were examined. The latest results of these observations will be shown.

1E1335 高速原子間力顕微鏡によるペルオキシレドキシン高分子量複合体の観察
Direct visualization of high molecular weight complex of peroxiredoxin using high-speed AFM

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Peroxiredoxin (Prx) is a ubiquitous peroxidase that reduces reactive oxygen species in the cell. Prx acts as not only a peroxidase but also a molecular chaperone, and the functional change is due to the formation of high molecular weight (HMW) complex by hyper-oxidation. In addition to hyper-oxidation, ATP/Mg also mediates the HMW complex formation of Prx. However, relationship between HMW complex formed by ATP/Mg and chaperone function is not clear. We therefore examined the structure of Prx HMW complex and its chaperone activity. High-speed AFM observation revealed that the structure of Prx HMW complex was spherical shape composed of rings. We will discuss the details of relationship between HMW complex formed by ATP/Mg and chaperone function.

1E1350 高速原子間力顕微鏡による Kai タンパク質間の相互作用の観察

High-speed AFM observation of dynamic interactions between Kai proteins

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The circadian rhythm in cyanobacteria is generated by an oscillator comprised of three Kai proteins (KaiA, KaiB and KaiC). KaiC consists of two hexameric rings (CI and CII) with a tail structure at the CII domain. It is known that interactions between three Kai proteins modulate the phosphorylation state of KaiC and determine the phase of circadian rhythm. However detailed molecular mechanism creating circadian rhythm in Kai system is still unclear. Structural analyses (EM, etc.) have suggested that both KaiA and KaiB bind to the CII ring, whereas a possibility of KaiB binding to the CI ring has also been proposed. Here we observe the interactions between KaiC and KaiA/B proteins using high-speed AFM to investigate the interaction dynamics.

1E1405 Development of structural analysis system of protein-protein complexes based on identification of hydrogen bond network

Masaru Tateno, Takuya Takeda, Jiyoung Kang (*Grad. Sch. Life Sci., Univ. Hyogo*)

Elucidation of the 3D structures of protein-protein (P-P) complexes is crucial to understand biological mechanisms at the atomic level. However, for P-P complexes with their weak/transient interactions, the crystallographic analyses are too difficult to determine the 3D structures. In this study, we developed a novel structural analysis system of P-P complexes, based on identification of interfacial hydrogen bond (HB) networks. We employed 32 crystal structures of the P-P complexes as a test set, and found that our system successfully identified ~94% of the P-P complex structures. In the session, the strategy of the flexible docking is also addressed, and the biological applications are discussed.

1E1425 効率よくウイルス DNA に変異を導入する APOBEC3G の脱アミノ化機構

The deamination mechanism of APOBEC3G required for effective gene mutation in viral genome

Keisuke Kamba[1,2], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Inst. of Advanced Energy, Kyoto Univ.*, [2]*Grad. Sch. of Energy Science, Kyoto Univ.*)

The human cytosine deaminase APOBEC3G (A3G) restricts HIV infection. A3G effectively deaminates the cytidines that are located close to the 5' end of the viral cDNA. However, the mechanism of the deamination reaction has not been elusive. In this study, we have investigated the recognition sequence and the deamination polarity of A3G by using the real-time NMR monitoring method that we developed previously. Firstly, we show that the 5 sequential nucleotides is the recognition sequence. Secondly, the electrostatic interactions between A3G and the phosphate backbone of an ssDNA is the key for sliding. Finally, we found that one of the HIV accessary proteins blocks sliding of A3G, thereby its activity.

1E1440 Structural modeling of negatively supercoiled DNA recognition peptide complexed with crossover DNA

Kakeru Sakabe, Jiyoung Kang, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

Lens Epithelium-derived Growth Factor (LEDGF)/p75 selectively binds to the negatively supercoiled DNA through the DNA-binding domain, where a cluster with the conservative amino acid sequence consisted of lysine and glutamic/aspartic acid residues is present. Surprisingly, even only this polypeptide segment extracted from the protein (e.g. $(Lys)_9(Glu)_9(Lys)_9$) exhibits the comparable selectivity for the negatively supercoiled DNA (Tsutsui, K., et al., NAR, 39 (2011), 5067-81). In this study, we conducted structural modeling of the peptide complexed with the spatially-crossing double stranded DNA, coupling to molecular dynamics (MD) simulations. We discuss the mechanisms of the selectivity based on the 3D structures of the complex.

1E1455 蛋白質の機能構造として働く励起構造

An excited-state conformer acts as the functional conformer of the protein

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The conformational fluctuations in the minimum DNA-binding domain of c-Myb, R2R3, were studied using CD and NMR techniques, under closely physiological conditions. A global unfolding transition takes place with a midpoint of transition around 50°C. In addition, the observation of simultaneous shift changes and broadening of NMR signals indicate the occurrence of locally fluctuating sub-state at physiological temperature. The locally fluctuating conformer is estimated to be present to some extent at 37°C, and is likely to be beneficial to the biological function, DNA-binding. This result is in agreement with the concept of an excited-state conformer that exists in equilibrium with the dominant ground-state conformer and acts as the functional conformer of the protein.

1E1510 過渡共鳴ラマン分光法を用いたプロテオロドプシン光反応初期中間体の解析

Photointermediates of proteorhodopsin studied by transient resonance Raman spectroscopy

Haruki Yamaryo[1], Shinichi Tajitsu[1], Jun Tamogami[2], Naoki Kamo[2], Masashi Unno[1] ([1]*Saga University*, [2]*Matsuyama University*)

Proteorhodopsin (PR) is a light-driven proton pump found in marine bacteria. Like a prototypical light-driven proton pump bacteriorhodopsin, PR contains a retinal chromophore covalently bound to a lysine residue via a protonated Schiff base linkage. In order to explore the early photointermediates, we have measured the transient resonance Raman (TRR) spectra of PR with 532 nm excitation. The TRR spectra of wild-type PR at 0.56, 2.2, and 5.4 ms resolutions are clearly different, indicating a presence of more than two photointermediates in this time range. We also performed a similar experiment for the E108Q mutant, where an N intermediate is absent. These results allow us to discuss a possible photocycle mechanism in PR.

1E1530 抗ニトロフェニル抗体の鍵となるアミノ酸残基 V_H33 の抗原結合における役割

Role of the key residue at V_H33 of anti-nitrophenyl antibody in its antigen binding

Yusui Sato[1], Takahiro Maruno[2], Harumi Fukada[3], Yuji Kobayashi[2], Takachika Azuma[4], Masayuki Oda[1] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ.*, [2]*Grad. Sch. of Eng., Osaka Univ.*, [3]*Grad. Sch. of Life and Environ. Sci., Osaka Pref. Univ.*, [4]*Res. Ins. for Biol. Sci., Tokyo Univ. of Sci.*)

During the course of affinity maturation of anti-nitrophenyl (NP) antibody, the W33L mutation could increase K_a of ~10-fold at the early stage of immunization. In this study, we prepared single-chain Fvs of germline N1G9 as well as highly matured C6, and introduced W33L mutation into these Fvs to elucidate the role of this mutation on the evolution of anti-NP antibodies. The experiments of antigen binding kinetics and thermodynamics showed that, upon the W33L mutation on N1G9, more favorable dissociation rate (k_{off}) and enthalpy change (ΔH) were observed, resulting in increasing K_a. On the other hand, upon the mutation on C6, less favorable k_{off} and ΔH, partially compensated by another parameter, were observed, resulting in decreasing K_a.

1E1545 抗体の新たな抗原認識機構がもたらす特異性創出原理

若手招待講演

New paradigm for antibody-antigen recognition enabling extraordinarily high specificity

Takamitsu Hattori[1], Darson Lai[1], Irina Dementieva[1], Sherwin Montano[1], Kohei Kurosawa[1], Akiko Koide[1], Alexander J. Ruthenburg[1,2], Shohei Koide[1] ([1]*Dept. of Biochem. and Mol. Biol., The Univ. of Chicago*, [2]*Dept. of Mol. Genetics and Cell Biol., The Univ. of Chicago*)

The antigen-binding site in the antigen-binding fragment of an antibody is the minimal and sufficient unit for the recognition of one antigen molecule. Here, we report antibodies that use two antigen-binding sites cooperatively to recognize an antigen. We isolated recombinant antibodies to histone methylation with high methylation-state and sequence specificities. Surprisingly, crystal structures and biophysical analyses revealed these antibodies form head-to-head dimers only when bound to antigens. This antigen clasping by two antigen-binding sites creates unusually large surfaces for antigen recognition with extreme specificity. This unexpected mechanism substantially broadens the paradigm of antibody-antigen recognition and will guide the design of new antibodies.

1E1600 Regulation of Adaptive Immunity: Activation and Inhibition of the ZAP-70 Kinase Domain

Roland G. Huber[1], Hao Fan[1,2], Peter J. Bond[1,2] ([1]*Bioinformatics Institute, A*STAR*, [2]*Department of Biological Sciences, NUS*)

We report microsecond timescale simulations of five distinct states of the ZAP-70 KD, comprising apo, inhibited and three phosphorylated variants. Extensive analysis of local flexibility and correlated motions reveal crucial transitions between the states, thus elucidating possible steps in the activation mechanism of the ZAP-70 KD. We explain inhibited complexes resembling an active-like conformation by showing that the inhibitor modulates the underlying protein dynamics and restricts it in a compact, rigid and inaccessible state to ligands or cofactors. Finally, our analysis reveals a novel, potentially druggable pocket in close proximity to the activation loop of the kinase, offering promise in targeting new pathways to specific kinase inhibition.

1J1320 ダイナミンによる膜切断メカニズムの高速 AFM イメージング解析

Pinch or Pop: HS-AFM imaging analyses of membrane scission mechanisms by Dynamin

Tetsuya Takeda[1,4], Yusuke Kumagai[2], Kaho Seyama[1], Huiran Yang[1], Hiroshi Yamada[1,4], Azuma Taoka[2,3], Takayuki Uchihashi[2,3,4], Kohji Takei[1,4], Toshio Ando[2,3,4] ([1]*Grad. Sch. Med. Dent. Pharma. Sci., Okayama Univ.*, [2]*Coll. Sci. Eng., Kanazawa Univ.*, [3]*Bio AFM, Kanazawa Univ.*, [4]*CREST, JST*)

Dynamin is a large GTPase responsible for membrane scission during endocytosis. Previous studies demonstrated that Dynamin forms a spiral around the neck of invaginating membrane. The Dynamin spiral severs membrane to form endocytic vesicles through GTP hydrolysis. Two mechanisms have been proposed for the membrane severing by Dynamin: “pinchase” model in which the spiral constricts to pinch off vesicles and “poppase” model in which the spiral extend lengthwise to push off vesicles. However, its precise mechanism remains to be elucidated.
In this study, we analysed Dynamin-mediated membrane scission by HS-AFM imaging of an in vitro reconstituted system. We will present up-to-date results about the structure and dynamics of Dynamin during membrane scission.

1J1335 Multi-state transitions of PTEN mediate spontaneous signal generation and environmental bias in cell migration

Satomi Matsuoka[1,2], Masahiro Ueda[1,2] ([1]*QBiC, Riken*, [2]*Grad. Sch. Sci., Osaka Univ.*)

PtdIns(3,4,5)P_3-enriched domain on cell membrane directs cell migration. We have examined single-molecule behaviors of 3-phosphatase, PTEN, in living *Dictyostelium discoideum* cells. PTEN undergoes transitions among 3 states with different lateral diffusivities. Enzymatically active state is the moderate one. Dephosphorylation accompanies prompt membrane dissociation, causing spontaneous domain formation via positive feedback. An amount of the active state depends on the transition from the slowest state. A chemoattractant suppresses this state, reducing the activity at the higher concentration side. It is illustrated that the molecular state transitions mediate the environmental bias upon the internal spontaneous signal shifting random into directed motility.

1J1350 Instantaneous fluorescence polarization microscopy for mapping position and orientation of protein assemblies in living cells with single molecule sensitivity

Tomomi Tani[1], Shalin Mehta[1], Molly McQuilken[2], Patricia Occhipinti[2], Amitabh Verma[1], Rudolf Oldenbourg[1], Amy Gladfelter[2] ([1]*Marine Biological Laboratory, USA*, [2]*Dartmouth College, USA*)

Dynamic molecular order underpins function of many cellular events. We report novel fluorescence polarization microscopy that analyzes position and orientation of live cellular assemblies with single-molecule precision at 10 Hz. By combining polarization-neutral excitation, instantaneous imaging along four polarization orientations and speckle image analysis, we study orientation of biomolecular assemblies in vitro and in living cells. We imaged dynamic orientation of actin filaments during rearward flow at the lamellipodia of keratinocytes. Filament orientation transitions from being orthogonal to the leading edge to parallel as the distance from leading edge increases. Our approach will be useful for mapping the 2D molecular orientation of variety of biomolecular assemblies in living cells.

1J1405 大気圧電子顕微鏡 ASEM による水中深さ方向の観察：神経の細胞輸送研究や組織の癌術中迅速診断への可能性

Depth observation of Tissues and cells in Liquid by ASEM: Applicability to Intra-Operative Cancer Diagnosis and cell trafficking study

Chikara Sato, Tatsuhiko Ebihara, Nassirhadjy Memtily, Mari Sato, Tomoko Okada, Masaaki Kawata (*AIST*)

ASEM images a 2- to 3-μm thickness of the sample resting on a silicon nitride-film window in the base of an open sample dish in liquid from below. In the present study, the specimen depths of labeled gold signals were determined using blurring of the dot using image analysis algorithm. Various mouse tissues (brain, spinal cord, muscle, heart, lung, liver, kidney, spleen and stomach) and neurons were fixed, stained with heavy metals, and visualized in radical scavenger D-glucose solution using the ASEM. Some stains made the nuclei of cells very prominent (platinum-blue, phosphotungstic acid). Notably, symbiotic bacteria were sometimes observed on stomach mucosa. Furthermore, kidney tissue could be stained and successfully imaged in

1J1425 癌進行に伴うヒト胃癌細胞の形状揺らぎと接着能の変化

Change in Shape Fluctuation and Adhesion of Human Gastric Cells Induced by Cancer Progression

Akihisa Yamamoto[1], Tatsuaki Tsuruyama[2], Motomu Tanaka[1,3] ([1]*iCeMS, Kyoto Univ.*, [2]*Diagn. Pathol., Kyoto Univ.*, [3]*Phys. Chem., Univ. of Heidelberg*)

The structure of multicellular tissues gets disordered according to the cancer progression. It is also known that single cells show a wide variety of size and shape (atypism), suggesting a decrease in connectivity. The main scopes of our study are (1) quantifying cellular morphology and (2) revealing the cell adhesion according to gastric cancer progression. Well-defined models of cell surfaces based on supported membranes [Tanaka & Sackmann, Nature (2005)] were used: (1) to quantify the adhesion force by pressure wave assay [Yoshikawa et al., JACS (2011)] as well as (2) to track the shape fluctuation by reflection interference contrast microscopy (RICM).

1J1440 １細胞分泌実時間イメージングが明らかにした細胞分泌動態の不均一性

Real-time single-cell secretion imaging revealed heterogeneity of the cell secretion

Yoshitaka Shirasaki[1,2], Ting Liu[3], Yoshifumi Yamaguchi[3], Mai Yamagishi[1,2], Nobutake Suzuki[1], Masayuki Miura[3], Osamu Ohara[2], Sotaro Uemura[1] ([1]*Grad. Sch. Sci., Tokyo Univ.*, [2]*IMS, RIKEN*, [3]*Grad. Sch. Pharm., Tokyo Univ.*)

Secretion of humoral factors, which mediate cell-cell communication, is a key process for maintaining homeostasis and regulating functions of the body. The secretory response of the cell is assumed under strict control depending on inputs. However, it is unclear how strictly regulated the secretory response is in individual cells.
We developed a platform for real-time single-cell secretion imaging by combining fluorescence immunoassay with the TIRF microscopy. On this platform, we clarified that the secretion dynamics of cytokines has different mode depending on kinds of cytokine. Furthermore, the secretory responses varied in quantity and in response time in individual cells though we traditionally assumed that they are uniform in same cell types.

1J1455 Visualizing mechanical force transmission at integrin molecules and the interior architecture of focal adhesions with traction maps

Masatoshi Morimatsu[1], Armen H. Mekhdjian[1], Alice C. Chang[1], Steven J. Tan[1], Alexander R. Dunn[1,2] ([1]*Department of Chemical Engineering, Stanford University*, [2]*Stanford Cardiovascular Institute, Stanford University School of Medicine*)

Mechanical interactions between cells and the extracellular matrix exert a profound influence on various cellular functions. However, how cells generate and detect mechanical force remains poorly understood. Here we describe an integrin-specific Forster resonance energy transfer -based molecular tension sensor (MTS) that allows us to directly visualize cellular traction forces. Simultaneous imaging of MTSs and GFP-tagged proteins results in maps of force-producing structures within focal adhesions with sub-diffraction spatial resolution. We find that αvβ3 integrin and Paxillin exhibit a high degree of spatial correlation with mechanical tension suggesting that these proteins may play direct roles in cellular mechanotransduction.

1J1510 細胞膜に繋留された小胞を介した新しいシグナル変換機構：1 分子イメジングによる解明

New signal transduction mechanism mediated by plasma-membrane-tethered vesicles: unraveling by single-molecule imaging

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Ligation-induced receptor activation is considered to trigger intracellular signaling generally by processes occurring in/on the plasma membrane (PM) and partially in endosomal membranes that contain activated receptors. Here, we found that, in immune mast cells, ~10% molecules of Linker for Activation of T cells (LAT), which is a major mediator for transferring the receptor activation signal to downstream signaling molecules, existed in cytoplasmic vesicles tethered to the PM via exocyst complexes for periods of 6 s or 73 s, which we termed LAT vesicles. Upon antigen stimulation, downstream signaling molecules were recruited to LAT vesicles. These results suggest that LAT vesicles, rather than LAT monomers or clusters in the PM, serve as key signaling platforms.

1J1530 GPCR の特徴であるリガンド無しでの構成的シグナルは過渡的な GPCR ダイマーが誘起している

Constitutive signaling without ligation characteristic with GPCRs is triggered by transient GPCR dimers

Rinshi Kasai[1,2], Akihiro Kusumi[1,2] ([1]*Inst. Front. Med. Sci., Kyoto Univ.*, [2]*WPI-iCeMS, Kyoto Univ.*)

Previously, we unequivocally showed that G-protein coupled receptors (GPCRs) are in dynamic equilibrium between monomers and dimers, with dimer lifetimes of ~100-ms. Nevertheless, the dimer function was unknown. Here, the functions of monomers and dimers were examined by observing single-molecule recruitment of trimeric G-proteins to GPCR's transient monomers and dimers, using a prototypical GPCR β2-adrenergic receptor. We found that trimeric Gs-proteins are recruited equally well to monomers and dimers, both before and after the agonist addition. However, the addition of inverse agonists, drugs blocking the GPCR's unique constitutive activity, inhibited Gs-protein recruitment to dimers. This indicates that GPCR dimers are responsible for GPCR's constitutive signals.

1J1545 **FlhA と FliH/FliI との相互作用がべん毛フックの構築順序を巧みに制御する**

Role of the interaction between FlhA and the FliH/FliI complex in coordinating flagellar hook assembly

Yumi Inoue[1], Miki Kinoshita[1], Keiichi Namba[1,2], **Tohru Minamino**[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*QBiC, RIKEN*)

The bacterial flagellar hook length is controlled at ca. 55 nm by an export switching device consisting of a molecular ruler FliK and an export switch FlhB. The FliH/FliI complex binds to export substrates in the cytoplasm and delivers them to the sorting platform made of nine copies of a membrane protein FlhA. We previously showed that the F459A mutation in FlhA affects the export of proteins needed for filament assembly but not that of hook-type substrates responsible for the structure and assembly of the hook, but it remains unknown how the FlhA platform coordinates protein export with assembly. In this study, we show that an interaction between FlhA and the FliH/FliI complex contributes to proper ordered export of hook-type substrates to control the hook length.

1J1600 **Biochemical and functional characterization of the effects of a single point mutation on mouse CP to CARMIL binding**

Ikuko Fujiwara[1], Christopher Alexander[2], Kirsten Remmert[2], Grzegorz Piszcek[2], John Hammer[2] ([1]*NITech*, [2]*NHLBI, NIH*)

To understand the regulatory mechanisms of CARMIL to CP (Capping Protein), a point mutation of CP, D44 of CP beta-subunit was employed. Bulk assays showed CP(b)D44N still strongly binds CAH3 domain of CARMIL, while CAH3 driven inhibition of CP(b)D44N was not detected. TIRF assay showed the actin elongation was paused for a long time even at low concentrations of the CP(b)D44N:CAH3 complex, caused by the slow dissociation of CP(b)D44N:CAH3 complexes from B-ends. Our data suggest the molecular dynamics of CP cannot be altered by CAH3 binding when D44 is missing, thus the CP(b)D44N:CAH3 complex still strongly caps B-ends.

1K1320 **高度高塩菌ハロバクテリウムサリナラムの遊泳運動特性の解析**

Characterization of the swimming motility of halophilic archaea, *Halobacterium salinarum*

Yoshiaki Kinosita[1], Nariya Uchida[2], Daisuke Nakane[1], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin University*, [2]*Department of Physics, Tohoku University*)

Archaea discovered at extreme conditions such as high temperature and high salt. Previous contributions showed that they swam by rotating archaeal flagella, but accurate quantification of rotation including the cell body was never clarified. Here, to formulate the swimming motility of archaea with physical parameters, we conducted following experiments on *Halobacterium salinarum*. (1) Characterization of the morphology of flagella under an electron microscopy. (2) Measurement of the rotational speed of flagella. Flagella were stained with a fluorescent probe and visualized by TIRF illumination. (3) Body rotation. Cells were labeled with QD605 and observed by 3-D tracking microscopy. Calculated velocity from the parameters provides a good agreement with the observation.

1K1335 **深海微生物の遊泳運動を高圧力下で観察する**

Direct observation of the swimming motility of deep-sea bacterium at high-pressure conditions

Masayoshi Nishiyama[1], Chiaki Kato[2], Yoshie Harada[3] ([1]*The HAKUBI Center, Kyoto Univ.*, [2]*JAMSTEC*, [3]*WPI-iCeMS, Kyoto Univ.*)

We have developed a high-pressure microscope that enables us to acquire high-resolution microscopic images. The developed system allowed us to study the bacterial motility at high-pressure conditions. Here, we monitored the swimming motility of *Schwanella violacea* strain DSS12 at optimum growth temperature of 8 °C. The strain was isolated from deep-sea sediment in the Ryukyu Trench at a depth of 5,110m. The swimming speeds at 0.1 and 50 MPa were about 20 and 16 μm s^{-1}, respectively. The speed of strain DSS12 was not largely changed by applied pressure, as compared with that that of *Escherichia coli* and *Vibrio alginoliticus* cells. Our results suggested that strain DSS12 is equipped with a motility machinery suitable for high hydrostatic pressure environments.

1K1350 **クラミドモナス鞭毛の波形の切り替え制御因子**

A regulation factor responsible for switching waveform of Chlamydomonas flagella

Junya Kirima[1], Misaki Shiraga[1], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst, NICT*)

The *Chlamydomonas* flagella show two types of waveform depending on intracellular Ca^{2+} concentrations. At pCa>6, wild-type falgella show asymmetric, ciliary-type waveform. On the other hand, the increase in $[Ca^{2+}]_i$ (5>pCa) elicits waveform change from asymmetric to symmetric, flagella-type waveform. Flagella of an outer-arm dynein (ODA) lacking mutant (*oda1*) show only asymmetric waveform even at high $[Ca^{2+}]_i$. Addition of wild-type crude ODA extract to *oda1* rescues the symmetrical waveform but high-purification of ODAs loses their ability to rescue *oda1* while keeps the ability to recover beat frequency of asymmetric waveform. The results suggest the presence of some factors for symmetric waveform. We have now found a novel protein with the EF-hand motif.

1K1405 **D32 のプロトン化で誘起されるべん毛モーター固定子 MotA/B の構造変化**

Structural change of the stator complex MotA/B on bacterial flagellar motor induced by protonation of D32

Yasutaka Nishihara[1], Akio Kitao[2] ([1]*Univ. of Tokyo, CMSI*, [2]*Univ. of Tokyo, IMCB*)

Bacterial flagellar motor consists of a rotor and ~10 stators. The stator is composed of 4 MotA and 2 MotB proteins for *Escherichia coli*. Although the torque is generated by proton binding at D32 on MotB, the molecular mechanism for the torque generation is still unclear.
To investigate the effect of D32 protonation, we performed the molecular modeling of the MotA/B complex and molecular dynamics simulations with protonated/non-protonated D32. Trajectory analysis shows that, for non-protonated D32, one helix in MotA was kinked at P173, whereas, for protonated D32, D32 formed a hydrogen bond with the main-chain O of D170 on MotA, which induced a straighter shape of the helix. This structural change seems to induce the structural change of the MotA cytoplasmic domain.

1K1425 Axonemal Dynein Light Chain-1 Locates at the Microtubule Binding Domain of the γ Heavy Chain

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The outer arm dynein (OAD) complex is the main propulsive force generator for ciliary/flagellar beating. Axonemal dynein light chain-1 (LC1) was found to be important for the ciliary/flagellar beating. However, its precise localization inside the OAD complex and its regulatory mechanism of OAD has not been unveiled.
Here, we performed Ni-NTA-nanogold labeling electron microscopy (EM), single particle analysis, and pull-down assays, and found that LC1 was associated with the microtubule binding domain (MTBD) of the γ heavy chain (HC). Together with observations that LC1 decreased the affinity of the γ MTBD for microtubules, we present a new model in which LC1 regulates OAD activity by modulating γ MTBD's affinity for the doublet microtubule.

1K1440 Off-axis motion of yeast cytoplasmic dynein takes a biased random walk

Mitsuhiro Sugawa[1], Shin Yamaguchi[1], Hiroaki Takagi[2], Keitaro Shibata[1,3], Yoko Y. Toyoshima[1], Junichiro Yajima[1] ([1]*Graduate School of Arts and Sciences, The University of Tokyo*, [2]*Department of Physics, Nara Medical University*, [3]*National Institute of Advanced Industrial Science and Technology*)

Yeast cytoplasmic dynein (CD) walks processively toward the minus end of a microtubule (MT). CDs also move laterally toward both the right and the left, switching randomly between the MT's protofilaments. But, the off-axis motion of each CD-coated bead is biased toward the left or the right and also fluctuates enough large to rotate occasionally around the MT. The distribution of the angular velocity is not gaussian but rather lognormal. These results suggest that the lateral motion of the CD-coated bead is a biased random walk which drives not only additive noise but also multiplicative noise. To reveal its mechanism, we are analyzing the temperature dependency of the biased random motion. We will discuss the results at the meeting.

1K1455 クライオ電子顕微鏡により明らかとなった微小管上を歩いている細胞質ダイニンの新規の構造と揺らぎ
Direct observation of cytoplasmic dynein stepping on microtubules by cryo-EM reveals a novel hinge at stalk-stalkhead junction

Hiroshi Imai[1,2], Tomohiro Shima[3], Kazuo Sutoh[4], Matthew L. Walker[5], Peter J. Knight[2], Takahide Kon[6], Stan A. Burgess[2] ([1]*Chuo Univ.*, [2]*Univ. of Leeds*, [3]*RIKEN QBiC*, [4]*Waseda Univ.*, [5]*MLW Consulting*, [6]*Grad. Sch. Sci., Osaka Univ.*)

Cytoplasmic dynein is an essential motor protein in human brain development by transporting cargoes in cells. The relative positions and the movement of the fluorescent labels on dynein, which was stepping on microtubules, have been reported. However, structure of dynein stepping on microtubules has been unknown. In order to know how dynein looks like when it steps on microtubules, we have directly observed Dictyostelium dynein dimers frozen while stepping along microtubules in the presence of Mg-ATP by cryo-electron microscopy. Two major structures of the motors were observed with novel intramolecular flexibility at the hinge between stalk and stalkhead (microtubule-binding domain). This hinge is unique to dynein among the motor proteins including kinesin and myosin.

1K1510 神経細胞オルガネラ輸送におけるキネシンとダイニンの数の測定：揺らぎの定理の応用
Measuring the numbers of kinesin and dynein on neuronal cargo transport using the fluctuation theorem

Kumiko Hayashi[1], Yasushi Okada[2] ([1]*Sch. Eng., Tohoku Univ.*, [2]*QBiC, RIKEN*)

We observed organelles, which are transported by kinesin and dynein in neurons, by using fluorescence microscopy. We investigated non-equilibrium fluctuation in the constant drag motion of the organelle's center position. We estimated the drag force acting on the organelle exerted by the motors using the fluctuation theorem, a theorem for entropy production that has been known in the field of non-equilibrium statistical mechanics. We found that the distribution of the drag force had several peaks. The values at these peaks may correspond to forces exerted cooperatively by multiple motors. The number of motors attached to an organelle is discussed based on the drag force distributions. We summarize our resent results on this issue.

1K1530 等方型 TRIFM とデフォーカスイメージングによる単一蛍光色素の角度と回転方向の検出
Detection of 3-D orientation and rotation handedness of single fluorophore by isotropic TIRFM and defocused imaging

Shoko Fujimura[1], Yuko Ito[2], Kengo Adachi[3], Mitsunori Ikeguchi[2], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin Univ.*, [2]*Medical Life Sci., Yokohama City Univ.*, [3]*Engin., Waseda Univ.*)

Determination of angle of the fluorophore at the single molecular level in an aqueous solution is crucial to uncover mechanochemical characteristics of enzymes such as motor proteins. We analyzed sequential defocused images of sparsely labelled microtubule that rotated on the lawns of single-headed kinesin under isotropic TIRFM (Adachi *et al.*, *Cell* 2007). Rotation was directly estimated with a precision of 1.4°, and the dipole angle against the MT axis distributed ~40-70°, which are consistent with results obtained from the Glide 6.4 XP scoring function and docking protocol. This methodology to detect 3-D orientation and rotating fluorophore will be widely applicable to detect domain motions of various proteins.

1K1545 １分子の複数状態モニタリングにより明らかとなった F_1-ATPase におけるヌクレオチド周辺の局所環境と化学状態の相関
Correlation between local environment around nucleotide and chemical state in F_1-ATPase revealed by single-molecule modes monitoring

Nagisa Mikami, Takayuki Nishizaka (*Dept. phys., Gakushuin Univ.*)

F_1-ATPase is a rotary motor powered by the chemical energy of ATP hydrolysis occurred at catalytic sites. The coupling between reaction sequence and shaft rotation has been challenged but not defined yet. Here we develop the polarization-modulation microscope that enables to quantify the intensity, mobility and orientation of a single fluorophore, and monitor the local environment around catalytic cores through fluorescent ATPs which directly correlates to the chemical state. The anisotropy of the fluorophore attaching to the bound nucleotide decreased from 0.6 to 0.4 by hydrolysis, indicating the partial opening of the catalytic site at the interface between α and β subunits. The nucleotide also rotated ～5° in CCW manner with the reaction.

1K1600 Single Molecule Time Series Analysis of F1-ATPase to Unravel the Role of Bound-ATP Hydrolysis

Chun-Biu Li, Tamiki Komatsuzaki (*RIES Hokkaido Univ.*)

F1-ATPase (F1) is the water-soluble component of the FoF1-ATP synthase that rotates upon ATP hydrolysis. Here I will report our recent studies of the possible roles of the bound-ATP hydrolysis reaction in terms of single F1 observations and time series analysis. We detect a small angular increment during the catalytic dwell triggered by the bound-ATP hydrolysis and find in free rotating F1 that the bound-ATP hydrolysis is followed by Pi-release with low synthesis rates. We then propose a functional role of the bound-ATP hydrolysis as a key to kinetically unlock the subsequent Pi-release in order to complete the catalytic cycle, despite its minor contributions to the torque and chemical energy generations in compared to the ATP binding and Pi-release.

1L1320 光センサータンパク質 TePixD の反応過程における過渡的揺らぎ

若手招待講演

Transient conformational fluctuation of TePixD during a reaction

Kunisato Kuroi[1], Koji Okajima[2,3], Masahiko Ikeuchi[2], Satoru Tokutomi[3], Masahide Terazima[4] ([1]*Inst. for Mol. Sci.*, [2]*Grad. Sch. Sci., Univ. Tokyo*, [3]*Grad. Sch. Sci., Univ. Osaka Pref.*, [4]*Grad. Sch. Sci., Univ. Kyoto*)

The conformational fluctuation of a protein is essential for functioning. Nowadays such fluctuation has been detected and attracted many interests. However it remains challenging to detect the fluctuation during a protein reaction for directly showing the relevance with functioning. In the present study, we developed the high-pressure transient grating (TG) system to detect the compressibility of transient intermediates during a reaction, since compressibility directly reflects the structural fluctuation. We applied this technique to the blue light sensor protein TePixD. TePixD has a unique decamer structure and dissociates to pentamers upon excitation. We succeeded in detecting its fluctuation during the photoreaction and showing its importance for the functioning.

1L1335 ラマン分光法による高濃度抗体溶液のタンパク質間相互作用の研究

The protein-protein interactions in highly concentrated antibody solution investigated by Raman spectroscopy

Chikashi Ota[1], Shintaro Noguchi[1], Kouhei Tsumoto[2,3,4] ([1]*Advanced R&D Center, Horiba, Ltd.*, [2]*School of Engineering, The University of Tokyo*, [3]*Institute of Medical Science, The University of Tokyo*, [4]*Drug Discovery Initiative, The University of Tokyo*)

In the biopharmaceutical industry, highly concentrated liquid formulations (>100 mg/ml) are required. To investigate protein-protein interaction of a highly concentrated antibody solution, the concentration dependence measurement has been carried out over a wide range of concentrations (10-200 mg/ml). The analysis of Raman bands of Amide I ,Tyr, Phe shows that in these wide range of concentrations, the secondary structure of the IgG molecules did not changed, however, the short range attractive interaction around Tyr and Phe started to work when the distance between molecules decreased as the size of IgG molecules. These conformational based knowledge by Raman spectroscopy will support the conventional colloidal approch, especially in the highly concentrated solution.

1L1350 Cys のチオレートと Trp のカチオン - π 相互作用による銅輸送タンパク質細胞外領域における一価銅安定化機構

Cysteine- and Tryptophan-Based Copper(I) Stabilization in the Extracellular N-terminal Domain of Ctr4

Mariko Okada, Takashi Miura, Takakazu Nakabayashi (*Grad. Sch. Pharm. Sci. Tohoku Univ.*)

The CXXSMXWNWYXXDXC motif with a symmetrical arrangement of Cys/Trp residues is conserved in extracellular N-terminal region of various fungi and yeast copper transporter proteins. We synthesized a peptide corresponding to this region of copper transporter Ctr4, and measured fluorescence, UV-visible absorption and Raman spectra in the presence and absence of copper. It is revealed that this domain readily reduces Cu2+ and traps copper ion through thiolate-Cu+ bond. This thiolate-Cu+ bond is stable even in air-saturated condition. The half-life of Cu+ bound by wild-type peptide is about 12 h, but is shortened to 8 h by tryptophan mutations. UVRR spectra show that the π-electrons of tryptophan indole ring may contribute to stabilization of the thiolate-Cu+.

1L1405 酸素の常磁性効果を利用したタンパク質の疎水性キャビティーの検出

Molecular oxygen as a paramagnetic NMR probe of dynamic hydrophobic cavity in proteins

Ryo Kitahara[1], Yuichi Yoshimura[2], Mengjun Xue[2], Frans A. A. Mulder[2] ([1]*College of Pharmaceutical Sciences, Ritsumeikan University*, [2]*Department of Chemistry and iNANO Center, University of Aarhus*)

Internal cavities in proteins are important structural elements that may facilitate functional motions. Here, we investigate binding of molecular oxygen (O2) into cavities in L99A mutant of T4 lysozyme by NMR spectroscopy. Upon increasing the O2 concentration to 8.9 mM (7 bar), changes in 1H, 15N, and 13C chemical shifts and signal broadening were observed for backbone amide and methyl groups located around two hydrophobic cavities of the protein. O2-induced longitudinal relaxation enhancements were measured for amide and methyl protons, and could be adequately accounted for by paramagnetic dipolar relaxation. The dissociation constant for O2 binding to the largest hydrophobic cavity was determined. O2 greatly prefers binding to hydrophobic over hydrophilic cavities.

1L1425 一分子蛍光顕微鏡による p53 変異体の標的配列探索ダイナミクスの観察

Observation of the Search Dynamics of p53 Mutants for the Target DNA Sequence by Single-molecule Fluorescence Microscopy

Yuji Itoh[1,2], Agato Murata[1,2], Seiji Sakamoto[1], Kei Nanatani[3], Takehiko Wada[1], Satoshi Takahashi[1,2], Kiyoto Kamagata[1,2] ([1]*IMRAM, Univ. Tohoku*, [2]*Grad. Sch. Sci., Univ. Tohoku*, [3]*Grad. Sch. Agr. Sci., Univ. Tohoku*)

The activated form of p53 slides along DNA, binds to the target DNA sequence, and promotes the expression of proteins required for tumor suppression. To understand the mechanism of the activity of p53, we constructed the pseudo-WT, activated, and inactive mutants of p53, and compared the search dynamics for the target sequence by single-molecule fluorescence microscopy. We observed the binding events to the target sequence and a significant numbers of the pass-through events over the target sequence. The binding probabilities (BP) of pseudo-WT, activated, and inactive mutants were respectively 10, 15, and 4%, indicating that BP is correlated with the activity of p53. p53 may regulate its function in a living cell by altering BP based on posttranslational modifications.

1L1440 DNA 結合蛋白質の単分子蛍光観察のための DNA 整列技術の開発

Development of a new method for making the array of aligned DNAs, DNA garden, for the single-molecule fluorescence imaging

Chihiro Igarashi[1,2], Agato Murata[1,2], Satoshi Takahashi[1,2], Kiyoto Kamagata[1,2] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. of Sci., Tohoku Univ.*)

A new method for making the array of aligned DNAs for single-molecule fluorescence measurements, DNA garden, was developed. The method is based on the microcontact printing of neutravidin on coverslip coated by MPC polymer and flowing biotinylated DNA. The protocol of DNA garden does not require microfabrication used in previous methods, which facilitates the single molecule investigation of the DNA and related proteins. We applied DNA garden to detect cleavage sites of restriction enzymes on DNA and to observe the sliding dynamics of a tumor suppressor p53 at a single-molecule level. Our results demonstrated the availability of DNA garden for the single-molecule functional assay of DNA binding proteins.

1L1455 X 線 1 分子追跡法・プローブ負荷試験による複合タンパク質・協同的運動の定量化

Quantification of Cooperative Motions for Multi-subunit Proteins by Single Molecule Loading test with Diffracted X-ray Tracking

Hiroshi Sekiguchi[1], Keigo Ikezaki[2], Naoto Yagi[1], Yuji Sasaki[1,2] ([1]*JASRI/SPring-8*, [2]*Grad. School Frontier Sci., Univ. Tokyo*)

Diffracted X-ray Tracking (DXT) is one of single molecule techniques for investigating intra-molecule dynamics of functional proteins. In DXT, a nanocrystal is immobilized on a target protein and the trajectory of its diffracted spot is analyzed as the motion of the protein.
The size of gold nanocrystal used for our measurements is ranged from 20 to 80 nm in diameter, and the size effect for the motion is not negligible. The angular motions for nicotinic acetylcholine receptor (nAChR) were depend on the size of gold nanocrystal (Sci. Rep. 4:6394 2014).
In this presentation, we review the relationship between the size of gold nanocrystal on the taraget protein, nAChR or group II chaperonin (PLoS ONE 8:e64176 2013), and the motion detected by DXT.

1L1510 Hydrolysis of lipid droplets by artificially designed peptides

Yoshihiro Iida, Atsuo Tamura (*Grad. Sci., Univ. Kobe*)

Obesity has become a huge problem in modern life. To avoid obesity, we should hydrolyze triglyceride in lipocyte. As a candidate for an anti-obesity drug, we tried to design peptides having enzyme activity like the lipase to hydrolyze triglyceride. The peptide was designed to have the alpha-helical conformation and catalytic triad composed of His, Asp and Ser. Based on this strategy, we synthesized peptides me1-5 and CPPme5. CD measurements showed that the conformations of all but me1 are alpha helical coiled-coils. Measurement of hydrolysis activity showed that me5 and CPPme5 had been conferred lipolysis activity. Based on these results, we conclude that the designed peptide can be regarded as a lipase mimic which is capable of hydrolyzing the lipid droplets.

1L1530 分子認識におけるメチル化の効果： 分子動力学計算による研究

Effect of methylation on molecular recognition: A molecular dynamics study

Takefumi Yamashita (*RCAST, Univ. Tokyo*)

Molecular recognition plays many important roles in a cell. A simple barometer of the recognition is the binding affinity. We sometimes experience that a single methylation influences the binding affinity several orders of magnitude more. In this study, we aim at clarifying the mechanism by which a methyl group enhances the binding affinity with an example system. We first performed molecular dynamics (MD) simulation-based binding free energy calculations both for a demethylated ligand and a methylated ligand with their target protein and confirmed these results are consistent with the experimental ones. Our analysis suggests that a single methylation enhance the binding affinity by significantly decreasing the dehydration free energy in this system.

1L1545 Investigating kinetics of conformational change using molecular dynamics and milestoning

Hiroshi Fujisaki[1], Ayori Mitsutake[2] ([1]*Nippon Medical School*, [2]*Keio Univ. Dep. Phys.*)

Large conformational change of biomolecules is vital for understanding their function but it takes much longer timescales than those of fluctuations around a basin, making it unfeasible to study using conventional molecular dynamics (MD) simulations. Because of recent advance of hardware, software, and novel algorithms, however, it has been possible to investigate the conformational change of relatively small biomolecular systems, and not only free energy landscape of biomolecules but also their kinetic networks can be clarified. Using MD simulations of chignolin at high temperature, we discuss how to analyze the MD data to extract kinetic information using milestoning, which is a simple and effective method to calculate the mean first passage times between milestones.

1L1600 自由エネルギーパスサンプリングへの PaCS-MD の応用

Application of PaCS-MD to Free Energy Path Sampling

Duy Tran[1], Akio Kitao[1,2] ([1]*Grad. Frontier Sci., Univ. of Tokyo*, [2]*IMCB, Univ. of Tokyo*)

Free energy calculation is of great interest for drug design because it is used to estimate binding affinity and rate constant. However, it is still not straightforward to accurately predict binding free energy because of accumulated errors in free energy calculation and unnatural dissociation pathway in reconstruction of the Potential of Mean Force. To overcome this barrier, we apply the Parallel Cascade Selection Molecular Dynamics (PaCS-MD) method to generate the conformational transition pathway without applying additional external biases. The binding free energy is extracted from the Umbrella Sampling using Weighted Histogram Analysis Method.

1M1320 Structural and functional comparison of hexahistidine tagged and untagged forms of small multidrug resistance protein, EmrE

Shahzada Junaid S. Qazi, Raymond Chew, Denice C. Bay, Raymond J. Turner (*Biological Science Department, University of Calgary, Alberta, Canada*)

We isolated EmrE using the two different purification methods, an organic solvent extraction method used to isolate UT-EmrE and nickel affinity chromatography of T-EmrE. All proteins were solubilized in the same buffered n-dodecyl-β-D-maltopyranoside (DDM) detergent and their conformations were examined in the presence/absence of different QCCs. The difference observed in in vitro and in vivo analysis will be presented that could be a result of the tag and/or the difference purification methods. We will present the data from both UT- and T-EmrE using biophysical techniques such as SDS-Tricine PAGE, dynamic light scattering and fluorescence spectroscopy for their functional and structural comparison.

1M1335 光駆動性ナトリウムイオンポンプによるナトリウムイオン輸送の構造基盤

若手招待講演

Structural basis for Na^+ transport mechanism by a light-driven Na^+ pump

Hideaki Kato[1], Keiichi Inoue[2], Rei Yoshizumi[2], Yoshitaka Kato[2], Hikaru Ono[2], Masae Konno[2], Shoko Hososhima[3], Toru Ishizuka[3], Mohammad R. Hoque[3], Hirofumi Kunitomo[4], Jumpei Ito[5], Susumu Yoshizawa[6], Keitaro Yamashita[7], Mizuki Takemoto[4], Tomohiro Nishizawa[4], Reiya taniguchi[4], Kazuhiro Kogure[6], Andres D. Maturana[5], Yuichi Iino[4], Hiromu Yawo[3], Ryuichiro Ishitani[4], Hideki Kandori[2], Osamu Nureki[4] ([1]*Sch. of Med., Stanford Univ.*, [2]*Grad. Sch. of Engineering, Nagoya Inst. of Tech.*, [3]*Grad. Sch. of Life Sci., Tohoku Univ.*, [4]*Grad. Sch. of Sci., Univ. of Tokyo*, [5]*Grad. Sch. of Bioagri. Sci., Nagoya Univ.*, [6]*Atmos. and Ocean Res. Inst., Univ. of Tokyo*, [7]*Harima Inst., Riken SPring-8*)

KR2 is the first light-driven Na^+ pump discovered. Since the positively charged Schiff base proton, located within the ion-transport pathway of all light-driven ion pumps, was thought to inhibit the transport of a non-proton cation, the discovery of KR2 raised the question of how it achieves Na^+ transport. Here we present KR2 structures in the resting and M-like intermediate states. Structural and spectroscopic analyses revealed the gating mechanism, whereby the flipping of Asp116 sequesters the Schiff base proton from the transport pathway. Together with the engineering of the first light-driven K^+ pumps and optogenetics experiments, our studies reveal the molecular basis for light-driven non-proton cation pumps and provide a framework for next-generation optogenetics.

1M1350 コヒーレントX線回折像から構造情報を抽出するための計算アルゴリズム

Computational algorithms to extract structural information from X-ray coherent diffractions

Atsushi Tokuhisa[1], Osamu Miyashita[1], Florence Tama[1,2] ([1]*RIKEN AICS*, [2]*Department of Physics, Nagoya University*)

Coherent diffraction patterns observed by X-ray free electron laser (XFEL) provide information on the biomolecular conformations. We are exploring new algorithms to model biomolecular structures from limited experimental data. Computational algorithms are used to generate hypothetical structural models that are in agreement with the experimental data. Toward this goal, we have estimated the XFEL beam intensity strength that is required to detect conformational differences observed in the biological systems with different sizes, such as a protein molecule and ribosome complex. We will also present new algorithms to quickly evaluate the agreement between the structural model and diffraction patterns using 1D X-ray diffraction intensity profile.

1M1405 X線自由電子レーザーによって明らかにされた光化学系II複合体の1.95Å分解能での無損傷構造

若手招待講演

Radiation damage free structure of oxygen evolving photosytem II at 1.95Å resolution revealed by X-ray Free Electron Laser

Michihiro Suga[1], Fusamichi Akita[1], Kunio Hirata[2], Go Ueno[2], Hironori Murakami[2], Yoshiki Nakajima[1], Tetsuya Shimizu[1], Keitaro Yamashita[2], Masaki Yamamoto[2], Hideo Ago[2], Jian-Ren Shen[1] ([1]*Okayama Univ.*, [2]*Riken Harima*)

The initial reaction of photosynthesis takes place in PSII, an enzyme which catalyzes photo-oxidation of water into dioxygen through an S-state cycle of the oxygen evolving complex (OEC). The structure of PSII has been solved by XRD at 1.9Å resolution, which revealed the OEC is a Mn4CaO5-cluster. However, EXAFS studies showed that the manganese atoms in the OEC are easily reduced by X-ray irradiation, and slight differences were found in the Mn-Mn distances between the results of XRD, EXAFS and theoretical studies. In this presentation, we will present a radiation-damage-free structure of PSII from Thermosynechococcus vulcanus in the S1 state at 1.95Å resolution using femtosecond X-ray pulses of SACLA and discuss water-splitting mechanism based on the structure.

1M1425 ファルネシル基結合型ヒトガレクチン1の構造

Structure of Human Galectin-1 Binding Farnesyl Group

Hirotsugu Hiramatsu, Kazumi Yamaguchi, Takakazu Nakabayashi (*Grad. Sch. Pharm. Sci., Tohoku Univ.*)

Human galectin-1 (hGal-1) that is known as a lectin to bind β-galactoside has an affinity to the farnesyl group. We analyzed the structure of hGal-1 in farnesyl thiosalicylic acid (FTS)-bound form. It was found that the environment of Trp68 in the FTS-bound form was more hydrophilic and the binding constant (KB) to lactose was smaller in comparison with the free hGal-1. Molecular dynamics simulation suggested that a loop including Trp68 becomes less flexible and the hydrogen bond with Asn50 is important to stabilize the structure in the FTS-bound form. The decrease in the flexibility of the loop brought about the hydrophilic environment of Trp68 in aqueous media. The Asn50-FTS interaction moved a few important residues from the sugar binding pocket and decreased KB.

1M1440 ADPリボシル化酵素C3とRhoA複合体の構造基盤

Structural basis of ADP-ribosyltransferase C3 exoenzyme with RhoA complex

Akiyuki Toda, Toshiharu Tsurumura, Toru Yoshida, Yayoi Tsumori, **Hideaki Tsuge** (*Kyoto Sangyo University*)

C3 exoenzyme is a mono-ADP-ribosyltransferase (ART) that catalyzes transfer of an ADP-ribose moiety from NAD+ to Rho GTPases. C3 has long been used to study the diverse regulatory functions of Rho GTPases. How C3 recognizes its substrate and ADP-ribosylation proceeds. Crystal structures of C3-RhoA complex reveal that C3 recognizes RhoA via switch regions. In C3-RhoA(GTP) and C3-RhoA(GDP), switch I and II adopt the GDP and GTP conformations, respectively, which explains why C3 can ADP-ribosylate both nucleotide forms. Based on structural information, we successfully changed Cdc42 to active substrate with combined mutations in the C3-Rho GTPase interface. The structures show directly for the first time that the ARTT-loop is the key to target protein recognition.

1M1455 ラン藻アナベナ由来 12 量体グルタミン合成酵素の結晶構造
Dodecameric crystal structure of Anabaena Glutamine synthetase

Waraphan Toniti[1], Toru Yoshida[1], Toshiharu Tsurumura[1], Hideaki Tsuge[1], Hiroyuki Ashida[2], Kayo Takahashi[2], Yoshihiro Sawa[2] ([1]*Kyoto Sangyo University*, [2]*Shimane University*)

Glutamine synthetase (GS) is an ATP-dependent enzyme. It plays roles in ammonia assimilation and glutamine biosynthesis. Anabaena sp. GS (AnaGS) was negative feedback controlled whereas E.coli GS was regulated by feedback inhibition and adenylylation by Adenylyltransferase (AT). According to the biochemical studies, some mutants AnaGS were adenylylated by EcoAT with almost no GS activity. This study aimed to get insight how adenylation inhibits the activity of GS in comparison between wild type and mutant AnaGS. We solved the structure by molecular replacement and accomplished the refinement. The structure showed hexamer in an asymmetric unit and the active dodecamer formed typical D6 symmetry similar to other GSs. We will discuss the regulation based on this structure.

1M1510 GFP の A および B state における高分解能 X 線結晶構造解析
X-ray crystallographic studies of GFP in the A and B states at high resolution

Kiyofumi Takaba, Kazuki Takeda, Kunio Miki (*Grad. Sch. Sci., Kyoto Univ.*)

The chromophore of green fluorescent protein (GFP) takes two forms in its structures, protonated “A” and deprotonated “B” forms. High-resolution structures including the protonation state of both forms are indispensable to discuss the relationships between the structure and the spectroscopic properties.
We used two GFP mutants, T203I and S65T, each of which have a structure of either A form or B form. Diffraction data were collected using synchrotron beam source under optimized conditions to suppress the radiation damage effect. We determined structures of these mutants at 1.30 Å and 0.95 Å resolution, respectively. These structures show slight but significant differences around the chromophore, which are related to the protonation state.

1M1530 中性子結晶構造解析によるセルロース加水分解酵素中のプロトン伝達経路の観測
Direct observation of proton pathway in cellulase by Neutron crystallography

Akihiko Nakamura[1], Takuya Ishida[2], Katuhiro Kusaka[3], Taro Yamada[3], Ichiro Tanaka[3], Nobuo Niimura[3], Masahiro Samejima[2], Kiyohiko Igarashi[2] ([1]*Okazaki Inst. for Integrative Bioscience*, [2]*Grad. Sch. Agr., Univ. Tokyo*, [3]*Univ. Ibaraki*)

Hydrolysis of carbohydrates is a major bio-reaction in nature, catalyzed by glycoside hydrolases (GHs). We employed neutron diffraction and high-resolution X-ray diffraction analyses to investigate the hydrogen-bond network in inverting cellulase PcCel45A, which is an endo-glucanase belonging to GH family 45, isolated from the mushroom Phanerochaete chrysosporium.
Examination of the enzyme and enzyme-ligand structures indicates a key role of multiple tautomerizations of asparagine residues and peptide bonds, which are finally connected to the catalytic residues, in forming the proton relay pathway of the catalytic cycle. Amide-imidic acid tautomerization of asparagine is necessary to reconsider interpretation of many enzymatic reactions.

1M1545 バクテリオファージ P22 の 2 次元結晶化
Two-dimensional crystallization of Bacteriophage P22

Hideyuki Yoshimura[1], Ethan Edwards[2], Dustin Patterson[2], Masaki Uchida[2], Kimberly McCoy[2], Rajarshi Roychoudhury[2], Benjamin Schwarz[2], Trevor Douglas[2] ([1]*Dep. Phys, Meiji Univ.*, [2]*Indiana Univ.*)

Here we report 2D crystallization of three different bacteriophage P22 morphologies: P22 procapsid (PC), enzyme encapsulated procapsids (β-glycosidase, enhanced green fluorescent protein, and NADH oxidase), empty shell (procapsid without scaffold proteins, ES), and the expanded form of P22 (EX). The 2D crystals of P22 VLPs were formed on a positively charged lipid monolayer at the water-air interface with a subphase containing 1% trehalose. The lipid monolayer with adsorbed P22 was transferred to a holey carbon grid and was examined by electron microscopy. The diffraction spots from the transferred film extended to the 6th order in stained samples and the 10th order in cryo-electron microscopy.

1M1600 Blender による生物学的アニメーションの制作と共有
Creating and shareing biological animations by Blender

Yutaka Ueno (*AIST Kansai*)

While molecular animations well demonstrate our biological findings and education, scientific community pays attention to neutral discussions if only laboratories that can afford computational burden for the production of such media. Since recently popularized computer graphics hardware encourages us for wider applications, we explored animation software Blender, a free software for three dimensional computer graphics modeling, for animating biological molecules. With examined methods for biological applications, the models for motility proteins such as actin-myosin and microtubule-kinesin are dedicated for biophysical discussions at our web site (http://cbrc.jp/~ueno/blend).

1N1320 生理的温度条件下での自律 DNA 計算に向けた DNA 生成反応システムの構築
Construction of a DNA generation reaction system for autonomous DNA-based computing at a physiological temperature

Ken Komiya, Kesu Dong, Toshio Takenaka, Masayuki Yamamura (*Interdisci. Grad. Sch. of Sci. & Engi., Tokyo Tech.*)

We had developed a DNA-responsive DNA state machine that can implement information processing via successive DNA binding and polymerization. Its computational program is encoded with the sequence of the component DNA strand. Such a DNA-based nanomachine for computational task is expected to be applied to control biological and medical processes. However, our machine requires sequential addition of short DNA strands as operational triggers for implementing each computational step at a physiological temperature. In the present study, we first report construction of a reaction system that generates distinct single-stranded DNA species in a programmed order. And then, we discuss integration of the proposed DNA generation reaction system with the DNA state machine.

1N1335 細胞折紙：3 次元共培養システムの構築
Cell origami technique for 3D cell co-culture system

Qian He, Takaharu Okajima, Kaori Shigetomi(Kuribayashi) (*Graduate School of Information Science and Technology of Hokkaido University*)

Cell co-culture system in 3D microenvironments like a human body is essential for investigating cellular functions.
Previously, we produced a 3D laden cell microstructure by using microelectromechanical systems (MEMS) and origami folding technology, which called cell origami. In this study, cell origami technique was applied to create a 3D cell co-culture system by using a combination of 3T3 and HepG2 cells. The result showed that the 3D cell co-culture system can be easily formed without any complicated process. The cell functions, such as albumin secretion and protein expression, will be detected in the future.
This system we developed may be useful for 3D co-culture of various types of cells.

1N1350 マイクロピラーによる細胞核の力学的拘束は正常細胞の増殖を抑制するが腫瘍細胞には影響しない
Mechanical trapping of the nucleus on microfabricated pillars inhibits the proliferation of normal cells but not tumor cells

Kazuaki Nagayama[1], Yumi Hamaji[2], Yuji Sato[2], Takeo Matsumoto[2] ([1]*Micro-Nano Biomechanics Laboratory, Department of Intelligent Systems Engineering, Ibaraki University*, [2]*Biomechanics Laboratory, Department of Mechanical Engineering, Nagoya Institute of Technology*)

We investigated the effects of nuclear deformation on cell proliferation using micropillared substrates. We found that the proliferation of vascular smooth muscle cells (SMCs) but not cervical cancer HeLa cells was dramatically inhibited on the micropillar substrates, even though remarkable nuclear deformation was observed in both types of cells. Mechanical testing with AFM revealed that SMC nuclei were over three times stiffer than those of HeLa, which consequently increased the nuclear mechanical resistance against extracellular microstructures. These results indicate that the proliferation inhibition resulted from deformation of the nucleus, which might be exposed to higher internal stress during nuclear deformation.

1N1405 血球細胞の流体力学的挙動の数値シミュレーション
A numerical study on the fluid dynamics of blood cells

Stephanie Nix[1], Yohsuke Imai[2], Takuji Ishikawa[2,3] ([1]*Fac. Sys. Sci. Tech., Akita Pref. Univ.*, [2]*Grad. Sch. Eng., Tohoku Univ.*, [3]*Grad. Sch. Biomed. Eng., Tohoku Univ.*)

In the microcirculation, red blood cells undergo axial migration, in which they are observed to move in the direction of the blood vessel axis. On the other hand, other cells found in the blood, such as white blood cells and platelets, are observed to flow primarily in the region near the blood vessel wall. In this study, we numerically investigate the influence of hydrodynamic factors on this difference in migration behavior. In particular, we analyze how the effects of the wall and flow curvature lead to a higher migration velocity for red blood cells compared to other cells found in the blood.

1N1425 機械受容チャネルによるコリネ型細菌のグルタミン酸放出機構の解析
Mechanism of glutamate export in Corynebacterium glutamicum through the mechanosensitive channel

Yoshitaka Nakayama[1], Kenjiro Yoshimura[2], Hidetoshi Iida[3], Hisashi Kawasaki[4], Reinhard Kraemer[5], Boris Martinac[1] ([1]*Victor Chang Cardiac Research Institute*, [2]*University of Maryland*, [3]*Tokyo Gakugei University*, [4]*Tokyo DENKI University*, [5]*University of Cologne*)

Corynebacterium glutamicum exports a large amount of glutamate through the mechanosensitive channel MscCG when the surface cell structure is changed. This process is used worldwide for the industrial glutamate production, and further improvement is required for the world demand. MscCG is a homolog of MscS, mechanosensitive channel functioning as an osmoregulator in E. coli, however they have remarkably structural and functional differences. In this study we investigate the gating characteristics of MscCG using microbial electrophysiological methods. MscCG has strong gating hysteresis due to slow kinetics and does not show the desensitization and inactivation mechanisms. These results suggest the possibility to improve the glutamate export in C. glutamicum.

1N1440 アンキリンリピートドメインと脂質の相互作用による TRPV1 チャネル活性の制御
Channel activity regulatory mechanism of TRPV1 by the interaction of ankyrin repeat domain with phospholipids

Kazuhiro Takemura[1], Shiro Suetsugu[2], Akio Kitao[1] ([1]*IMCB, Univ. Tokyo*, [2]*Grad. Sch. Biol. Sci., NAIST*)

The transient receptor potential subfamiliy V (TRPV) is a non-selective cation channel activated by a various stimuli. A recent study showed that the channel activity of TRPV4 was modulated by direct interactions of the ankyrin repeat domain (ARD) with phospholipids in the membrane. In this study we conducted molecular dynamics simulation of TRPV1 tetramer in the membrane. We constructed three models of the tetramer in the membrane, namely, open, partially open, and close which describe the state of the channel. We observed the large flexible motion of TRPV1 tetramer which enhance the interaction of ARD with phospholipid in the membrane. How the observed interactions affect the channels in TRPV1 will be discussed.

1N1455 真正細菌のポンプ型ロドプシンの機能転換およびそれらの光反応についての研究
Functional conversion of eubacterial pump rhodopsins and the investigation of the photoreactions

Yurika Nomura[1], Keiichi Inoue[1,2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*PRESTO*)

Light-driven ion pump rhodopsins are classified into proton-, chloride- and sodium pump, whose.characteristic motifs are DTD(E), NTQ and NDQ, respectively. These motifs must be important for their functions, but, there could be other residues that determine their functional differences. Here, we attempted the functional conversion of pump by mutations of the motifs and additional residues to identify critical amino acids for each function of eubacterial pumps. As the result, some mutants succesfully converted functions, whereas the others did not. Then, photoreactions of these mutants were studied. We'll discuss molecular mechanism of successful and unsuccessful functional conversions.

1N1510 膜輸送体の超高感度活性計測のための新しいマイクロデバイス

Novel micro device for highly sensitive measurement of membrane transporter activities

Rikiya Watanabe[1,2], Naoki Soga[1], Hiroyuki Noji[1] ([1]*Department of Applied Chemistry, The University of Tokyo*, [2]*PRESTO, JST*)

We present a novel micro device (el-ALBiC) that displays a sub-million of lipid bilayer chambers, each equipped with nano-size electrodes. Because nano-size electrodes enable a modulation of membrane voltage, the el-ALBiC performed the highly sensitive detection of voltage-driven membrane transporter activity, i.e., proton pumping by F_oF_1-ATP synthase. Thus, the novel micro device, el-ALBiC, largely extended the versatility of arrayed lipid bilayer chamber system, and holds promise for further analytical and pharmacological applications such as drug screening.

1N1530 Mechanism of sodium/proton antiporter from transition path simulations

Kei-ichi Okazaki, Judith Warnau, Gerhard Hummer (*Max Planck Institute of Biophysics*)

Na^+/H^+ antiporters are secondary active transporters, important for controlling pH, salt concentration, and volume of the cells. Recently, crystallographic structures of NhaP in the inward-open state from *Pyrococcus abyssi* (PaNhaP) and *Methanocaldococcus jannaschii* (MjNhaP) have been solved. Also, low-resolution structure of MjNhaP in the outward-open state was obtained by electron cryo-crystallography. Since PaNhaP and MjNhaP are close homolog, these structures give us an opportunity to investigate the transport cycle. Here, we model the atomistic structure of the outward-open state from the EM map by a flexible fitting algorithm. Then, we obtain a natural transition path between these two states by a transition path sampling technique.

1N1545 Molecular mechanisms of proton transfer in H^+-coupled MATE in outward facing form

Wataru Nishima[1], Wataru Mizukami[2], Yoshiki Tanaka[3], Ryuichiro Ishitani[4], Osamu Nureki[4], Yuji Sugita[1] ([1]*RIKEN Theoretical Molecular Science Laboratory*, [2]*Department of Material Sciences, Faculty of Engineering Sciences, Kyushu University*, [3]*Laboratory of Membrane Molecular Biology, the Graduate School of Biological Sciences, Nara Institute of Science and Technology*, [4]*Department of Biophysics and Biochemistry, the University of Tokyo*)

MATE (the Multidrug And Toxic compound Extrusion) is one of the five multidrug transporter super-families. It utilizes H^+ gradient across membrane to transport lipophilic and cationic substrates. Recently, structures of the MATE from *P.furiosus* (PfMATE), has been determined by X-ray crystallography in outward-facing form. Two distinct structures *Straight* and *Bent* are characterized by conformations of TM1, indicating D41 and D184 are H^+ binding sites.

To elucidate the molecular mechanism of H^+ transfer in MATE, we performed MD and QM/MM simulations of PfMATE. Simulation results suggest that D41 is protonated in *Straight*, while D41 and D184 are both protonated in *Bent*. The protonation of D184 is also suggested to drive the conformational change from *Straight* to *Bent*.

1N1600 Multiscale Dynamics of Flaviviridae Fusion Peptides: Membrane Interactions via Simulation and Experiment

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In this work, a multiscale molecular dynamics simulation approach was employed to investigate the mechanisms of Falviviruses fusion peptides (FP) interacting with lipid bilayers. Atomic-resolution free energy profiles were calculated as a function of insertion within membranes. These profiles were correlated with peptide structural and lipid-peptide interaction data, and validated by fluorescence spectroscopy experiments. W101A muatnt was found to possess ~50% of the wild type FP affinity. The binding affinity was reduced significantly for the wild-type FP interaction with pure POPC membranes in comparison with realistic anionic endosomal lipids. Herein, novel insights into the structural and thermodynamic bases for the Flavivirus infection process have been gained.

1O1320 転写因子 Ets1 天然変性領域のリン酸化による DNA 認識阻害メカニズムの検討

Molecular Mechanism of Inhibition of Transcription Factor Ets1-DNA Binding, Induced by Phosphorylation on a Disordered Region

Kota Kasahara[1], Junichi Higo[1], Masaaki Shiina[2], Kazuhiro Ogata[2], Haruki Nakamura[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Med., Yokohama City Univ.*)

It is well known that phosphorelation at an intrinsically disordered region (IDR) of a transcription factor Ets1 regulates its DNA binding. However, its molecular mechanism was still unclear. We performed molecular dynamics simulations with exhaustive conformational sampling of the IDR to compute the free energy landscapes by using our own method, V-McMD (Higo et al., J. Comput. Chem. in press). Consequently, the phosphorylated IDR forms several different stable conformations at 300 K, each of which is attracted by positive charges on the recognition helix (H3) and competitively inhibits DNA specific binding. This can be so called "Multistery" of IDR. On the contrary, in the case of non-phosphorylated IDR, the probability of the direct interactions to H3 is very low.

1O1335 光活性化 bZIP モジュール Photozipper の DNA 結合性

Evaluation of DNA-binding of a light-activatable bZIP module, Photozipper

Osamu Hisatomi, Yuki Yabe, Yoichi Nakatani (*Grad. Sch. of Sci., Osaka Univ.*)

Photozipper (PZ) is an engineered light-regulated basic leucine zipper (bZIP) module containing a bZIP domain and a light-oxygen-voltage-sensing (LOV) domain of aureochrome-1. We investigated DNA-binding of PZ by several techniques including size exclusion chromatography (SEC) and fluorescent correlation spectroscopy (FCS). SEC analysis demonstrated that the light-induced dimerization of PZ reversibly enhanced its affinity for the target DNA. FCS data suggested that the dissociation constant of PZ dimer/DNA complex in the light state was in the tens of nanomolar range in the presence of 140 mM KCl. PZ may be used as a light-activatable bZIP module to control the assembly of biomolecules in the cell.

1O1350 3 本鎖 DNA 結合蛋白質の 3 本鎖 DNA 認識機構
Molecular mechanism of triplex DNA-binding proteins to recognize triplex DNA

Hidetaka Torigoe, Kazuki Kiuchi, Kikue Mase, Norihiro Sato, Takuma Katayama (*Department of Applied Chemistry, Faculty of Science, Tokyo University of Science*)

Budding yeast Stm1 and human U2AF65 are triplex DNA-binding proteins. Here, we examined the molecular mechanism of the triplex DNA-binding proteins to recognize triplex DNA. We found that Stm1(1-113 aa) and U2AF65(1-98 aa) were the minimal domains to bind to triplex DNA. The ability of the triplex binding domains (TBDs) to bind to each of the triplexes containing a series of point mutations or quite different base sequences was quite similar in magnitude to that to the triplex without any mutations. The TBDs may recognize the shape rather than the base sequence of the triplex. CD spectra showed that the TBDs were intrinsically disordered regions. The conformational flexibility may enable the TBDs to recognize any triplex sequences with the similar binding ability.

1O1405 HU Binding Coupled Bending of Double Stranded DNA

Cheng Tan, Tsuyoshi Terakawa, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

HU, one of the most abundant DNA architectural proteins, binds DNA preferentially to the intrinsically curved regions and stabilizes the bending of DNA. We use coarse-grained MD simulations to investigate the coupling between HU binding and conformational changes of DNA. We find that HU scans for the favorable binding sites on DNA in a rotation uncoupled manner. While having only slight preference for the AT-rich sequence, HU binds to nicks or gaps with significantly high probabilities. Our results show that the binding of HU facilitates the bending of DNA and stabilizes the bent structure, which is consistent with previous experimental results. We also reveal the effect of ion concentration on the binding of HU and the conformational changes of the HU-DNA complex.

1O1425 リバースジャイレースによる DNA の二重らせんをきつく巻きつける反応の解析
Direct observation of DNA overwinding by reverse gyrase

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Reverse gyrase, found in hyperthermophiles, is the only enzyme known to overwind DNA. Here, we image the reaction at 71 °C under a microscope. A single reverse gyrase molecule processively winds the DNA for >100 turns. The unloaded reaction rate exceeds 5 turns/s, which is >100-fold higher than hitherto indicated but lower than the ATPase rate of 20 1/s, indicating loose coupling. The overwinding reaction sharply slows down as the torsional stress accumulates in DNA and ceases at stress of mere ~5 pN·nm. The enzyme would thus keep DNA in a slightly overwound state to protect the genome of hyperthermophiles against thermal melting. All results point to the mechanism where strand passage relying on thermal motions, as in topoisomerase IA, is biased toward overwinding.

1O1440 Single-molecule measurement for sliding dynamics of tumor suppressor p53 on DNA

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1D sliding of a tumor suppressor p53 along DNA is an essential dynamics for its effective search of the target sequence on DNA genome. Assuming that the function of p53 might be affected by intranuclear environment, we investigated 1D sliding of p53 at different concentrations of divalent cations. Based on the single-molecule fluorescence microscopy, we revealed that the diffusion constants of p53 increase significantly at the higher [Mg2+] and [Ca2+]. The dissociation rate constants of p53 from DNA also become faster at the higher [Mg2+] and [Ca2+]. Thus, while the 1D-sliding of p53 is dependent on [Mg2+] and [Ca2+], the sliding distance of p53 per one binding event to DNA is maintained nearly constant.

1O1455 Direct visualization of *E.coli* SbcD enzymatic activity on single strand DNA by high-speed AFM

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E.coli SbcD is primarily a Mn(II)-dependent endonuclease for single strand DNA (ssDNA). Herein, we studied dynamic events associated with the enzymatic activity of SbcD on ssDNA using high-speed AFM. While ssDNA was being degraded by SbcD, globules were formed gradually, which finally grew to large globules consisting of DNA and a large number of SbcD molecules. This globule formation occurred in a Mn(II)-dependent manner. This surprising result perhaps indicates that SbcD possesses an additional activity to assemble ssDNA. This study enriches our knowledge of proteinaceous enzyme-based DNA metabolism and could provide new insights into the molecular process of nuclease activity.

1O1510 高速 AFM が捉えた大腸菌 MukB の構造動態
Structural dynamics of *E. coil* MukB captured by HS-AFM

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MukB is a structural maintenance of chromosomes (SMC) protein that is involved in chromosome segregation and condensation in *E. coli*. Although its detailed static structure is already known, its structural dynamics and functional mechanism have remained elusive. Here, we used high-speed AFM to reveal the dynamic structure and action of MukB at the single-molecule level. We first observed MukB alone to characterize its overall structure including head, coiled-coil and hinge domains as well as their dynamic variations. Then, we studied the impact on the MukB's structure and behavior of ATP, DNA and partners, MukE and MukF. In the presentation, we will report obtained results in detail.

1O1530 Structural dynamics of tri-nucleosome by coarse-grained simulations: effects of histone acetylation

Le Chang, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

Histone acetylation, which neutralizes the charge of specific lysine side-chain, has been proved to be able to activate gene expression. Experimental finding indicates that acetylation of H4 histone N-terminal tail could induce decompaction of chromatin fibre. However, the structural detail of decompacted chromatin and its relationship to transcription are still unclear. In this work, we studied a system consist of three nucleosomes by coarse-grained simulations with CafeMol. The simulations have different setups including H3, H4, H2A, H2B acetylated at lysine residues in N-terminal tail as well as one without acetylation. It is found that the acetylation of H4 is most effective for structural decompaction.

1O1545 粗視化分子シミュレーションによるクロマチン環境下における転写因子と ERK の拡散運動の研究
The diffusion dynamics of transcription factors and ERK in chromatin environment studied by coarse-grained simulation

Ryo Kanada[1], Tsuyoshi Terakawa[1], Hiroo Kenzaki[2], Shoji Takada[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*ACCC, RIKEN*)

The transcription factors (TFs) and ERK protein (which is in the signal transduction pathway) support the biological function by regulating the gene expression and the activity of transcription. The regulation mechanics of the gene expression is assumed to be qualitatively related to the dynamical behavior of TFs in the intra-nuclear crowded environment. However the details of the dynamics of TFs and ERK in the nucleus are still unknown in experiments. So, in this work, we investigated the diffusion dynamics of ERK and the representative TFs such as p53, HMGB1, and Oct4 in the chromatin structure that is composed of 20 nucleosomes densely by conducting coarse-grained simulation with CafeMol software. The detail comparison of the diffusion between TFs would be reported.

1O1600 粗視化シミュレーションから見えてきたクロマチン凝縮ダイナミクスにおける H1 C 末端変性部位の重要性
Disordered tail of the linker histone H1 in chromatin compaction dynamics studied by coarse-grained simulations

Nobu C. Shirai, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

Linker histone H1 binds to nucleosomes and induces more compact chromatin structures. Although recent experiments detected the binding sites of H1 globular domain in nucleosomes, it is rather difficult to detect the position of disordered C-terminal tail of H1. Here, we performed coarse-grained molecular dynamics simulations of H1 with nucleosomes to investigate effects of the disordered tail on the compaction dynamics of chromatin. We observed that the H1 winds its disordered tail around the linker DNA of the nucleosome and induces compact chromatin structures through several compaction pathways. These characteristic compaction processes suggested the importance of the disordered tail of H1.

1Q1320 固液界面における二成分系脂肪酸単分子膜の研究
Study on structural formation of mixed fatty acid monolayers at liquid/solid interface

Masahiro Hibino[1], Yoshitsugu Mukaiyama[2] ([1]*Dept. Appl. Sci., Muroran Inst. Tech.*, [2]*Toyota Motor*)

Self-assembled monolayers (SAM) of binary mixtures of fatty acids adsorbed on a graphite surface were studied using scanning tunneling microscopy (STM) at the liquid/solid interface. The STM images of the SAMs show the entire area is filled with short and long bright bands corresponding to the alkyl chains of two kinds of fatty acids. The long fatty acid molecules are adsorbed preferentially, that is, the high concentration of short molecules at which the domain originated from the short molecules grows sharply is required. To understand the adsorption mechanism in mixed fatty acids at the interface, the difference of free energy per methyl group is estimated on the basis of the observed STM images.

1Q1335 飽和リン脂質／コレステロール二成分混合膜系における相挙動の詳細解析
Detailed phase behavior analysis in the saturated 1,2-diacylphosphatidylcholine/cholesterol binary monolayer system

Tsubasa Miyoshi, Satoru Kato (*Grad. Sch. Sci & Tech., Univ. Kwansei Gakuin*)

The surface pressure-area isotherms of DMPC, DPPC, or DSPC (diacyl PC)/cholesterol binary monolayers were measured with high accuracy to get insight into the lateral molecular packing in these binary systems. Addition of cholesterol induces the formation of a highly condensed phase where the diacyl PC molecule has an surface area even smaller than that in the solid phase, irrespective of the surface pressure and the hydrocarbon chain length. Moreover, in order to explain the phase behaviors semi-quantitatively, we introduced “vicinity lipids” surrounding a cholesterol molecule in the low cholesterol concentration region and proposed a simple model, which is fundamentally consistent with the phase behavior of the binary monolayer system.

1Q1350 シミュレーションを用いた脂質二重膜の相転移の理論的研究
A theoretical study of thermal phase transition of phospholipid bilayer

Koji Ogata, Shinichiro Nakamura (*RIKEN Innovation Center*)

We performed all-atom molecular-dynamics (MD) simulations to investigate the thermal phase behavior of two hydrated DPPC and DPPE. The structures of DPPC in the crystalline (L_c), gel (L_β), ripple (P_β) and liquid crystalline (L_α) phases and those of DPPE in the L_c and L_α phases were identified from the trajectories in the MD simulations. We proposed the mechanism of the structural transformation from L_β to P_β. However, the main phase transition temperature (T_m) values showed errors ~50 K from the experimentally measured values. To reduce these errors, the parameters for vdW were modified to fit the T_m values of the simulation to the experimental values. After the fitting procedure, the T_m values of the lipids improved, and the errors of T_m improved from ~50 K to ~15 K.

1Q1405 ER 膜タンパク質の膜貫通配列のリン脂質 flip-flop を促進する物理化学的性質の解析

Physicochemical properties of membrane-spanning sequences in the ER proteins to promote phospholipid flip-flop

Hiroyuki Nakao[1], Keisuke Ikeda[2], Yasushi Ishihama[1], Minoru Nakano[2] ([1]*Grad. Sch. Pharm. Sci., Kyoto Univ.*, [2]*Grad. Sch. Med. Pharm. Sci., Univ. Toyama*)

Phospholipid flip-flop in the endoplasmic reticulum (ER) is very rapid in order to maintain lipid homeostasis in the membrane. However, proteins responsible for the flip-flop are unknown. We have previously demonstrated that a hydrophilic residue in the transmembrane region is effective to promote flip-flop. Therefore, we hypothesized that ER proteins witch have hydrophilic residues in the transmembrane region may promote flip-flop. In this study, we synthesized peptides with native membrane-spanning sequences of the proteins and investigated their flip-flop promotion ability. We found the sequence to increase flip-flop rate at a very low peptide/lipid ratio, and revealed the physicochemical property required for the promotion of flip-flop.

1Q1425 シトクロム P450 基質薬剤クロルゾキサゾンのリン脂質 POPC 膜結合に対するコレステロールの阻害効果の X 線回折による研究

X-ray diffraction studies of the effect of cholesterol on the binding of cytochrome P450 substrate drug chlorzoxazone to POPC bilayers

Ayumi Yamada[1], Nobutaka Shimizu[2], Takaaki Hikima[3], Masaki Takata[3,4], Toshihide Kobayashi[5], **Hiroshi Takahashi**[1,3,5] ([1]*Grad. Sch. Sci & Tech., Gunma Univ.*, [2]*KEK-PF*, [3]*Harima Inst., Riken*, [4]*IMRAM, Tohoku Univ.*, [5]*Riken*)

In the last year, we reported that the presence of cholesterol affects the binding of chlorzoxazone (CZX), which is a muscle relaxant drug and is a substrate of liver-specific cytochrome P450 (CYP), to phosphatidylcholine (PC) bilayers. The aim of the present study was to investigate the cholesterol content dependence of the binding-inhibition effect by the use of X-ray diffraction technique. Our analysis using model electron density profiles suggested that high cholesterol content (30-50 mol%) tends to prevent CZX from binding to POPC bilayers, and that, on the other hand, low cholesterol content (about 10 mol%) shows an opposite effect. We will discuss the role of cholesterol on the drug metabolism associated with CYP.

1Q1440 脂質ナノディスクを形成する膜活性ポリマー

Membrane-active amphiphilic polymers for lipid bilayer nanodisc formation

Kazuma Yasuhara, Jin Arakida, Jun-ichi Kikuchi (*Grad. Sch. Mat. Sci., Nara Inst. Sci. Tech.*)

We have investigated the fabrication of lipid bilayer nanodiscs by designed membrane-active amphiphilic polymers to provide a prospective molecular platform for membrane protein solubilization. We synthesized a variety of amphiphilic polymethacrylate random copolymers. Characterization of obtaind lipid-polymer complex was performed by means of dynamic light scattering, TEM observation and differential scanning Calorimetry. It was found that the formation lipid bilayer nanodiscs depended on the structure of polymers such as hydrophilic group, the amphiphilic balance as well as the molecular weight. We expect that this technique can be applied to the direct fragmentation of cell membrane to achieve detergent-free reconstitution of membrane proteins.

1Q1455 ナノ粒子の細胞内取り込み機構：膜変形と粒子拡散

Physical mechanism of cellular uptake of nanoparticles: membrane deformation and diffusion of adsorption particles

Kazuki Shigyou, Ken Nagai, Tsutomu Hamada (*JAIST Material*)

In the past decade, nanoparticles have been reported to show cellular toxicity. However, biophysical mechanism of the toxicity, such as uptake process of nanoparticles into a cell, is still unclear. In this study, we used an artificial cell, such as cell-seized liposomes, to clarify the physical mechanism of the interaction between nano/submicro-particles and lipid bilayer membranes. First, we focused on the membrane deformation induced by particle adsorption. Then, we measured the lateral diffusion of particles on the membrane surface.

1Q1510 バナナ状タンパク質による膜チューブ形成

Membrane tubulation induced by banana-shaped proteins

Hiroshi Noguchi (*ISSP, Univ. Tokyo*)

In living cells, shape deformations of biomembranes are controlled by various proteins. Many of these proteins contain a binding module known as the BAR (Bin-Amphiphysin-Rvs) domain, which consists of a banana-shaped dimer. We have studied how anisotropic spontaneous curvatures of banana-shaped domains induce effective interaction between proteins and change membrane shapes using implicit-solvent meshless membrane simulations. Our study reveals that a small spontaneous curvature perpendicular to the rod can remarkably alter the tubulation dynamics at high protein density whereas minor effects are only obtained at low density. A percolated network, which suppresses tubule protrusion, is intermediately formed for negative perpendicular curvatures.

1Q1530 電場に駆動される自発運動ゲル

Self-propelled gel particles driven by electric field

Ken Nagai[1], Masayuki Hayakawa[2], Masahiro Takinoue[2,3] ([1]*JAIST*, [2]*Tokyo Tech.*, [3]*JST*)

Since the similar collective motions are observed in the groups of various living things, it is believed that there exist universality classes for collective motions. Actually, Solon et al. reported that the collective motion of self-propelled particles highly depends on the symmetry of the motion of each particle using simple agent-based models. To investigate experimentally the dependence of collective motion on the symmetry and the shape of the particles, we developed self-propelled gel particles using the Centrifuge-based droplet shooting device (CDSD). We can create gel particles with various types of symmetry and shape with CDSD. We will report the analysis of the self-propelled motion of the particles driven by the uniform AC electric field.

1Q1545 概日時計における周期の頑健性と位相の可塑性の互恵的関係
Reciprocity between robustness of period and plasticity of phase in biological clocks

Tetsuhiro S. Hatakeyama, Kunihiko Kaneko (*The University of Tokyo, Department of Basic Science*)

Circadian clocks exhibit the robustness of period and plasticity of phase against environmental changes such as temperature and nutrient conditions. Thus far, however, it is unclear how both are simultaneously achieved. By investigating distinct models of circadian clocks, we demonstrate reciprocity between robustness and plasticity: higher robustness in the period implies higher plasticity in the phase, where changes in period and in phase follow a linear relationship with a negative coefficient. The robustness of period is achieved by the adaptation on the limit cycle via a concentration change of a buffer molecule, whose temporal change leads to a phase shift. Universality of reciprocity is confirmed with an analysis of simple models.

1Q1600 フィードバックループのあるシグナル伝達におけるマックスウェルのデーモン
若手招待講演

Maxwell's demon in biochemical signal transduction with feedback loop

Sosuke Ito[1], Takahiro Sagawa[2] ([1]*Department of Physics, Tokyo Institute of Technology*, [2]*Department of Applied Physics, the University of Tokyo*)

Unified theory of information and thermodynamics has been intensively discussed for nonequilibrium stochastic systems from a viewpoint of a thought experiment known as Maxwell’s demon. Recent information-thermodynamic studies reveal that the second law of thermodynamics can be generalized for a broad class of nonequilbrium dynamics including biochemical autonomous systems. In this study, we apply results of information-thermodynamic studies of Maxwell's demon to biochemical signal transduction. We focus on the negative feedback loop in sensory adaptation, e.g., E. coli biochemical chemotaxis, and show that the generalized second law of thermodynamics reveals a fundamental informational bound of the robustness of adaptation against environmental noise.

2B1355 アミロイド線維表面構造に対するヨウ素プローブの利用
Utilizing iodine as a probe for surface structures of amyloid fibrils

Takato Hiramatsu, Seongmin Ha, Yuki Masuda, Eri Chatani (*Grad. Sch. Sci., Kobe Univ.*)

Amyloid fibrils are implicated in many diseases such as Alzheimer's. Despite of apparent structural similarities, they often exhibit polymorphism bearing various degrees of cytotoxicity. In this study, we focus on iodine staining as a new method for distinguishing structural differences of fibrils. When three types of fibrils made of insulin were stained with KI/I2 solution, iodine molecules adsorbed onto the fibril surfaces and showed clearly different absorption spectra in visible region. Each spectral shape was conserved even after seeding reaction. As a result of detailed analysis of iodine titration, at least three specific sites with distinct binding constant were clarified, suggesting that iodine staining can probe detailed surface structures of amyloid fibrils.

2B1410 テラヘルツ時間領域分光および誘電分光を用いたアミロイド線維水和水ダイナミクスの観測
Hydration Water Dynamics of Amyloid Fibrils Studied by Terahertz Time-domain Spectroscopy and Dielectric Spectroscopy

Naoki Yamamoto[1], Keisuke Tominaga[1,2], Eri Chatani[1] ([1]*Graduate School of Science, Kobe University*, [2]*Molecular Photoscience Research Center, Kobe University*)

Amyloid fibrils have been known to relate to some serious diseases like Alzheimer disease. Because the fibril formation is achieved in the thermal fluctuation of the solvent, water, it is important to understand how hydration water dynamics relates to the fibril formation mechanisms. In this study we observed temperature dependence of hydration water dynamics of amyloid fibrils made from human insulin by using terahertz time-domain spectroscopy and dielectric spectroscopy. We found that the relaxation time of hydration water of the amyloid fibril, which was at around 15 picoseconds at 293 K, was faster than that of native insulin, indicating that the surface of the amyloid fibrils retains different physicochemical properties from that of native globular proteins.

2B1425 NMR による天然変性蛋白質の残存構造解析
Residual structures in the intrinsically disordered proteins monitored by NMR

Chiaki Nishimura (*Fac. Pharm. Sci., Teikyo Heisei Univ.*)

For two intrinsically disordered proteins, the residual structure was elucidated by NMR. The comparative studies were conducted on the C-terminal segment of peripherin-2 and measles virus Ntail domain. In both proteins, only a helical structure was observed in the middle parts of proteins based on the chemical shift. Furthermore, the faint beta-structure was detected for two proteins by the delta-2D analyses. The amide-proton exchange was monitored by CLEANEX-PM method. Different from the location of helix, the significant protection was observed at the N-terminus for periherin-2 and C-terminal part of helix for Ntail. The estimation of the content of the residual structure will be discussed compared to those of the other proteins and in the folding intermediate.

2B1440 蛋白質高エネルギー構造パラダイムの役割
The role of the "high-energy paradigm" of protein structure

Kazuyuki Akasaka (*Kyoto Pref. Univ., Grad. Sch., Bio. Envir. Sci.*)

Protein molecules are intrinsically dynamic in functioning. A powerful new tool for detecting the fluctuation is the high-pressure NMR spectroscopy introduced by the author's group [1]. The spectroscopy detects fluctuations in a wide conformation space [2] through atomic-detailed structural changes in NMR under variable pressure [3], involving cooperative motions of atoms into "high-energy sub-states" in the so-called "high-energy paradigm", crucial for function [4,5].

1. Akasaka K, Yamada H. (2001) Methods Enzym 338:134-58.
2. Akasaka K (2006) Chem Rev 106(5):1814-35.
3. Hirata F, Akasaka K (2015): J. Chem. Phys. 142(4):044110.
4. Fourme R, Girard E, Akasaka K (2012): Curr Opin Struct Biol 22(5): 636-642.
5. Akasaka K (2014): High Pressure Research 34(2):222-235.

2B1455 シアノバクテリア由来アルカン合成関連酵素の機能解析及びバイオエネルギー生産への応用
Functional analysis of the cyanobacterial enzyme for alkane biosynthesis and its application for bioenergy production

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Cyanobacteria synthesize a small amount of hydrocarbons that can be used as bioenergy. This reaction is catalyzed by acyl-(acyl carrier protein) reductase (AAR) and aldehyde deformylating oxygenase (AD). Because AAR has very low catalytic activity, it is necessary to improve its activity for bioalkane production. Here, we revealed that AAR from *Synechococcus elongatus* PCC 7942 has the highest activity among AARs from various cyanobacteria. Mutational analysis to search for the determinants of the AAR activity is ongoing. Finally, we transformed the cyanobacterium *Synechocystis* sp. PCC 6803 with the genes of the most active AAR and AD. As a result, we have succeeded in photosynthetic production of alkanes/alkenes with higher amount than the pseudo-wild-type strain.

2B1515 Accelerated H-DROP: An SVM based Helical Domain linker pRedictor trained with OPtimized features

Richa Tambi, Soichiro Ide, Ryosuke Suzuki, Teppei Ebina, Yutaka Kuroda (*Tokyo University of Agriculture and Technology*)

Domain linker prediction is the focus of much interest as it can help identifying novel domains suitable for high throughput proteomics analysis. Here, we report an accelerated version of SVM-based helical linker predictor, H-DROP. H-DROP is, to the best of our knowledge, the first predictor for specifically and effectively identifying helical linkers. The prediction sensitivity and precision of the original H-DROP were 35.2% and 38.8%, respectively, which are at least 10% higher than those of previous methods. We have now reduced the calculation time of H-DROP from about 10 minutes per sequence to ~30 seconds with sensitivity and precision of 33.3% and 36.4% , respectively. H-DROP is freely available from http://domserv.lab.tuat.ac.jp/

2B1530 活性化システインを持つ蛋白質構造の理論設計

Theoretical Design of Protein Scaffold Harboring an Activated Cysteine

Takahiro Kosugi, David Baker (*Dept. of Biochem., Univ. Washington*)

Many artificial enzymes to date have been reported because they are interesting and important for both academic research and industry. However, these enzymes have designed based on naturally occurring scaffolds and the scaffolds limits the sites that can be designed and can result in unexpected structural rearrangements. In this study, we designed a protein scaffold to put an activated cysteine residue whose activity is supported by histidine residue. The designed protein is expressed and stable, and following sequence optimization rapidly reacts with an active site probe in a manner dependent on the catalytic dyad residues. Moreover, E coli cells expressing the design showed much higher reactivity to a fluorescent active site probe than non-expressing cells.

2B1545 RE3Volutionary Computational Design of symmetric proteins

Arnout Voet (*Yokohama, RIKEN CLST*)

The modular nature of the protein architectures suggest that proteins have evolved by the duplication and fusion of genetic fragments to give rise to modular, often symmetric protein architectures. We have developed a computational protein design workshow in which we REverse Engineer Evolution to create symmetrically self assembling functional protein building blocks.
Our computational protein design workflow has been successful for the design of different proteins. Using these proteins we have demonstrated mathematical guided selfassembly according to the lowest common multiplier rule. Redesigned symmetric proteins are able to biomineralize nano crystals resulting in protein dimerization, and other proteins are suitable for cage formation.

2C0845 金ナノロッドを用いたキネシン頭部構造変化の観察による協調的二足歩行の仕組みの解明

Direct Observation of the Allosteric Conformational Change of Kinesin-1 using Gold Nanorod and its Implication for Head-head Coordination

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Kinesin-1 is a motor protein that moves along microtubules in a hand-over-hand manner. The tension posed to the neck linker has been shown to be essential for the coordination, however the structural basis is still unknown. Here, we observed the rotational motion associated with ATP isomerization in the motor domain (head) by observing orientation of single gold nanorod attached to the head using high-speed dark-field microscopy. We show that the subdomain rotation is suppressed in the leading position, where the neck linker is prohibited from docking, and is stabilized in the trailing position after ATP binding followed by the neck linker docking. These results explain how the neck linker tension allosterically regulates ATPase cycle in the leading and trailing heads.

2C0900 ミオシンの協調的首振りとアクチン滑り運動のゆらぎ

Cooperative lever-arm swings of myosins and fluctuation of actin sliding

Yota Kondo, Kazuo Sasaki (*Dept. Appl. Phys., Sch. Eng., Univ. Tohoku*)

Myosin II is a motor protein which swings a lever arm and generates actin sliding. If the actions of individual myosins are independent, the effective diffusion coefficient D of actin sliding was predicted to be proportional to the inverse of the actin length L (K. Sekimoto and K. Tawada, Biophys. Chem., 2001). However, a measurement of actin sliding *in vitro* showed that D is almost independent of L (N. Noda *et al*. Biophysics, 2005). To explain the experiment, we analyzed the myosin cooperation by using a simplified Duke model and found that the D - L relation depends on the number of bound myosins as well as the myosin cooperation. In order to verify this result, we carry out computer simulation of actin sliding by using the original Duke model.

2C0915 プログラム可能な DNA バネにより明らかにされたミオシン VI の外力依存的なギアチェンジ機構

Force-induced Gear-change Mechanism of Myosin VI Revealed with Programmed DNA Origami Spring

Mitsuhiro Iwaki[1,2], Shelley Wickham[3,4,5], Keigo Ikezaki[6], Toshio Yanagida[1,2,7], William Shih[3,4,5] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*, [3]*Dana-Farber Cancer Inst.*, [4]*Harvard Med. Sch.*, [5]*Wyss Inst. Harvard Univ.*, [6]*Univ. of Tokyo*, [7]*CiNet*)

Mechanical stimuli initiate physiological processes that regulate diverse cellular behavior. The transduction of these stimuli includes mechano-sensitive components such as motor proteins, ion channels and cytoskeletal filaments. There exist few methodologies that can measure mechano-responses at the molecular level, however. Here, we developed a programmed spring module (nanospring) using DNA origami to monitor the mechano-response of human myosin VI, a motor protein that has a role in hearing, at the single molecule level. Single molecule nanoimaging of fluorescently labeled myosin VI tethered to a nanospring showed force-induced switching between transporting and anchoring on actin filaments by application of hand-over-hand and inchworm mechanisms.

2C0930 ダイニンとアクチン結合タンパク質を基に新しいモータータンパク質をデザインする

Designing novel biomolecular motors based on dynein and actin-binding proteins

Akane Furuta, Kazuhiro Oiwa, Hiroaki Kojima, **Ken'ya Furuta** (*Bio ICT Lab, NICT*)

The mechanism of molecular motors have been studied in a top-down manner where the molecules are divided into pieces and analysed. However, critical factors for a protein machine to produce directional movement are still unclear. Here, we adopt a bottom-up strategy to identify the critical factors through the reconstruction of a molecular motor. We show that synthetic motors--combinations of a motor core of microtubule-based dynein and non-motor actin-binding proteins--actually drive the movement of an actin filament. Moreover, the direction of actin movement can be reversed simply by changing the relative position of the motor core and the actin binding module. Our strategy provides the basic design principle of a molecular motor for designing controllable biomachines.

2C0945 マイコプラズマ・モービレの滑走装置に関わる新奇タンパク質

Novel protein involving gliding machinery of *Mycoplasma mobile*

Tasuku Hamaguchi, Yuhei O. Tahara, Daiki Matsuike, Makoto Miyata (*Grad. Sch. of Sci. Osaka City Univ.*)

Mycoplasma mobile, a pathogenic bacteria of fresh water fish, glides on solid surfaces smoothly using ATP energy. The gliding machinery is composed of the large surface proteins and the internal structure composed of dozens of genes including F_oF_1-ATPase paralogs. In this study, we focused on MMOB1650 which may bridge between internal and surface structures. MMOB1650 has one and two transmembrane segments near N- and C-termini, respectively. This protein was purified as a complex with Gli521, a force transmitter on the cell surface. We observed the molecular shape of this complex by rotary shadowing electron microscopy, and found an additional mass at the center of Gli521 triskelion, which was reported previously.

2C1355 テロメア繰り返し配列含有 RNA の生細胞内 1 分子イメージング

Single molecule imaging of telomeric repeat-containinng RNA in living cells

Hideaki Yoshimura, Toshimichi Yamada, Hiroki Segawa, Takeaki Ozawa (*Sch. Sci., Univ. Tokyo*)

In this study, we target to reveal the functional mechanism of telomere-repeat containing RNA (TERRA). TERRA is transcribed from telomere and is suggested to have functions such as transportation of telomere-related proteins and stabilization of RNA-protein complexes on telomeres. Analysis of TERRA dynamics in living cells will provide clues to understand the mechanisms of TERRA functions. Here, we developed a fluorescent probe to visualize TERRA in living cells and performed single molecule imaging of TERRA in living cells. In this experiment, we found that some diffusing TERRA molecules transiently confined around a telomere. From the results of single molecule analysis of TERRA motions, the TERRA functions in living cells will be discussed.

2C1410 Three dimensional trafficking of membrane protein PAR-1, labelled with quantum dot carried by endocytotic vesicles

Seohyun Lee[1], Motoshi Kaya[1], Kohsuke Gonda[2], Hideo Higuchi[1] ([1]*Graduate School of Science, the University of Tokyo*, [2]*Graduate School of Medicine, Tohoku Univeristy*)

Our research is to visualize the endocytotic movement of PAR-1 carrying vesicles in three dimension, and investigate the effect of PAR-1 activation on its movement using thrombin. PAR-1 is one of the G-protein coupled membrane protein involved in the signal pathway which results in increasing cell mobility. Thrombin is the agonist of PAR-1 activation by cleaving its N-terminal while initiating endocytosis of PAR-1. Labelling with quantum dot, we observed the endocytotic movement of PAR-1 carrying vesicles in three dimension using 3D microscopy. As a result, characteristic trafficking movements of PAR-1 are tracked and activated PAR-1 by thrombin showed higher diffusion velocity compared to non-activated PAR-1.

2C1425 長時間１蛍光分子観察法の開発と応用：インテグリンの動的架橋が細胞接着を担う

Development of long-term single fluorescent-molecule tracking revealed dynamic integrin crossbridging for cell adhesion

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Photobleaching and photoblinkng observed in single fluorescent-molecule imaging were suppressed by including molecular oxygen and both oxidizing and reducing reagents in the specimen. Single integrin molecules could be tracked for >100 s without blinking. This allowed us to find that integrin molecules rapidly diffuse even in the focal adhesion (FA), with intermittent stationary periods lasting for <0.5, 6, and 40 s (exponential lifetimes), perhaps for responding to external force applied at various loading rates. Long stationary events often occurred in zones in the FA where stress is highest. These results suggest that dynamic, rather than stable, binding of integrins to actin and the extracellular matrix are responsible for cell adhesion and its regulation.

2C1440 A fast- and positively photoswitchable fluorescent protein for ultralow-laser-power RESOLFT nanoscopy

K Dhermendra Tiwari[1], Yoshiyuki Arai[1], Masahito Yamanaka[2], Tomoki Matsuda[1], Masakazu Agetsuma[1], Masahiro Nakano[1], Katsumasa Fujita[2], **Takeharu Nagai**[1] ([1]*ISIR, Osaka Univ.*, [2]*Dept. Appl. Phys., Osaka Univ.*)

Fluorescence nanoscopy has revolutionized our ability to visualize biological structures not resolvable by conventional microscopy. However, photodamage induced by intense light exposure has limited its use in live specimens. Here we describe Kohinoor, a fast-switching, positively photoswitchable fluorescent protein, and show that it has high photostability over many switching repeats. With Kohinoor, we achieved super-resolution imaging of live HeLa cells using biocompatible, ultralow laser intensity (0.004 J/cm^2) in reversible saturable optical fluorescence transition (RESOLFT) nanoscopy.

2C1515 高速光スイッチング蛍光タンパク質の蛍光偏光変調・励起角狭帯化照明による生体に優しい超解像イメージング

Biocompatible superresolution imaging by polarization demodulation/excitation angle narrowing of fast photoswitching fluorescent proteins

Tetsuichi Wazawa, Hiroki Takauchi, Tiwari Dhermendra, Yoshiyuki Arai, Tomoki Matsuda, Takeharu Nagai (*ISIR, Osaka U*)

Nanoscopy of fluorophores by polarization demodulation/excitation polarization angle narrowing (SPoD-ExPAN) could perform fast acquisition (>30 Hz) of images with high spatial resolution (<50 nm). Availability of conventional fluorophores including GFP is one of the advantage in this method whereas intense light illumination (> MW/cm^2) used for stimulated emission depletion of excited fluorophore in ExPAN would cause damage on live specimens . To eliminate this drawback, we used a positively photoswitching fluorescent protein (PAFP), Kohinoor, which has the fastest switching on/off property among the PSFPs reported to date. Accordantly, we could dramatically reduce the power density of illumination light (5 W/cm^2) allowing to apply low power light sources such as LED.

2C1530 フェムト秒ファイバーレーザーで励起可能な高効率二光子蛍光分子

Efficient Two-Photon Fluorescent Molecules Excitable by a Femtosecond Fiber Laser

Yasutaka Suzuki[1], Hiroki Moritomo[1], Akinari Fuji[1], Takato Shiraishi[2] ([1]*Graduate School of Medicine, Yamaguchi University*, [2]*Faculty of Science*)

Femtosecond fiber laser is a low cost and user-friendly light source for two-photon (TP) excitation. The laser typically emits femtosecond pulse at ~1000 nm. However, fluorescent molecules excitable at this wavelength region are very limited. In the present study, we developed efficient TP molecules excitable by a femtosecond fiber laser and accumulated on mitochondria. By using the molecules, we observed two-photon fluorescence images of mitochondria of the Hek293 cell using a fiber laser operating at 1030 nm. In addition, TP molecules exhibiting fluorescence within a wavelength region so called the 'tissue optical window' were developed. These molecules are promising candidates for realizing deep tissue imaging.

2C1545 複数の光操作と組み合わせた膜電位メージングと薬剤スクリーニングの可能性を広げる発光指示薬の開発

Luminescent indicator expands application for functional voltage imaging with multiple optical manipulation and drug screening

Shigenori Inagaki[1], Tomoki Matsuda[1], Yoshiyuki Arai[1], Yuka Jinno[2], Hidekazu Tsutsui[2,3], Yasushi Okamura[2] ([1]*ISIR., Univ. Osaka*, [2]*Grad. Sch. Med., Univ. Osaka*, [3]*Sch. Mat. Sci., JAIST*)

Luminescence imaging which doesn't require extrinsic light source has advantages over fluorescence imaging especially in terms of its complete compatibility with optogenetic tools and long-term observation. Here, we introduce a world-first luminescent indicator for voltage phenomena, LOTUS-V. LOTUS-V successfully visualizes voltage change triggered by channelrhodpsin2 and halorhodpsin in the single cell. Moreover, LOTUS-V is applicable into monitoring drug effect onto cardiomyocytes derived from human induced pluripotent stem cell (hiPSC-CM) by long-term imaging. Voltage imaging by LOTUS-V opens the door to explore detail investigations into phenomena regarding membrane voltage that is left behind by fluorescence imaging.

2C1600 マルチカラー・リアルタイム生物発光イメージングのための３色の超高輝度発光タンパク質 Nano-lantern の開発

Multicolor Nano-lanterns: the tricolored and super-brilliant luminescent proteins for multicolor, real-time bioluminescence imaging

Akira Takai[1], Masahiro Nakano[2], Kenta Saito[2], Remi Haruno[2], Tomonobu M. Watanabe[1,3], Tatsuya Ohyanagi[1], Takashi Jin[1], Yasushi Okada[1], Takeharu Nagai[1,2,3] ([1]*QBiC, RIKEN*, [2]*ISIR, Osaka Univ.*, [3]*PRESTO, JST*)

The application of luminescence imaging has been limited mainly by the two drawbacks of luciferases: low brightness and poor color variants. Here, we report the development of cyan and orange luminescent proteins approximately 20 times brighter than the wild-type luciferase (Takai et al., PNAS 2015). The color change and enhanced brightness were both achieved by exploring bioluminescence resonance energy transfer (BRET) from luciferases to fluorescent proteins, a technology that we previously reported for the development of the bright yellowish-green luminescent protein Nano-lantern. These tricolored multicolor Nano-lanterns enable monitoring of multiple cellular events, including dynamics of subcellular structures, gene expressions, and intracellular Ca^{2+} change.

2D1355 FTIR 分光法を用いたイエロープロテインの発色団周囲の水素結合環境の解析

Analysis of the hydrogen-bonding envirinment around the chromophore of photoactive yellow protein by FTIR spectroscopy

Tatsuya Iwata[1,2], Hideki Kandori[1,2] ([1]*Dept. Eng., NITech*, [2]*OBtRC, NITech*)

Photoactive yellow protein (PYP) is a blue light receptor found in purple bacteria. One of debates has been the presence of a low barrier hydrogen bond (LBHB) between anionized *p*-coumaric acid (pCA) chromophore and the O-H group of Glu46. To investigate hydrogen-bonding environment around pCA in PYP, light-induced difference FTIR spectroscopy was applied. Measurements of unlabeled, isotope-labeled PYP and mutants enabled us to assign the FTIR signal of the O-H stretch of Glu46. We observed no isotope effect of pCA-labeled on the O-H stretch of Glu46, excluding proton sharing under our experimental conditions. Present FTIR study does not support LBHB between pCA and Glu46, though the hydrogen bond of Glu46 is very strong.

2D1410 MALDI-TOF-MS を用いた BLUF ドメインの同位体標識の解析

Analysis of isotopic labeling of BLUF domain using MALDI-TOF-MS

Takashi Nagai[1], Tatsuya Iwata[1], Shota Ito[1], Mineo Iseki[2], Masakatsu Watanabe[3], Shinya Kitagawa[1], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Toho University*, [3]*The Graduate School for the Creation of New Photonics Industries*)

BLUF (sensor of blue-light using FAD) domain is a photoreceptor that binds flavin (FAD) as a chromophore. Unlike other flavin-binding photoreceptors, the chemical structure of FAD does not change at all between unphotolyzed and light-activated intermediate (BLUFred) states. Conserved Tyr and Gln residues are prerequisite for function and keto-enol tautomerization of Gln has been proposed. However, enol form of Gln was not observed clearly by FTIR spectroscopy of isotope-labeled protein. We now examine isotope-labeling of Gln using MALDI-TOF-MS, and the mechanism of BLUF activation will be discussed based on these results.

2D1425 青色光センサー蛋白質 BlrP1 の光反応ダイナミクス

Photo-induced reaction dynamics of blue light sensory protein BlrP1

Kousei Shibata, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci., Univ. Kyoto*)

Blrp1 is a bacterial blue light sensor protein consisting of a light sensing BLUF domain and an enzymatic EAL domain which hydrolyses c-di-GMP. In order to understand the signal transduction mechanism, we investigated the reaction dynamics of full-length BlrP1 and shorter construct containing only BLUF domain by the transient grating method. Upon photoexcitation, the diffusion coefficient (D) of full-length BlrP1 decreased dramatically, indicating higher order structure changed during the photoreaction. When the shorter construct was photoexcited, however, the D change did not take place and only a small volume change was observed. These results suggest that the small conformational change in BLUF domain transmit the light signal to the EAL domain for its activation.

2D1440 光反応検出によって見えてくるフォトトロピンの多様性
Diversity of phototropin studied from the viewpoint of photoreaction dynamics

Yusuke Nakasone[1], Koji Okajima[2], Yusuke Aihara[1], Akira Nagatani[1], Satoru Tokutomi[2], Masahide Terazima[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Osaka Prefecture Univ.*)

Phototropins are blue-light dependent kinases in plants and green algae. They contain two LOV domains as light sensing modules and the reaction dynamics of them have been studied by the transient grating method. We have found that the reactions of LOV domains from Chlamydomonas reinharditii (Cr) are totally different from those of LOV domains from Arabidopsis thaliana (At). In the case of Cr-phot, the LOV1 and its C-terminal helices showed drastic diffusion coefficient change upon photoexcitation, whereas the LOV2 did not undergo global reaction. This observation was contrary to the case of At-phot, in which the excitation of LOV2 resulted in the unfolding of its C-terminal helices. These findings suggest the phototropins have diversity in their signaling mechanism.

2D1455 遺伝子改変した LOV タンパク質中の FMN の光還元反応
Photochemical reactions of FMN in the mutant LOV proteins

Nozomi Ueda, Yukiko Ono, Tatsuya Iwata, Masayo Iwaki, Hideki Kandori (*Nagoya Institute of Technology*)

The LOV domain functions as a light sensor of phototropins, which binds FMN as a chromophore. Light irradiation generates FMN*, followed by the adduct formation between FMN and the nearby Cys residue. Here, we examined the possibility that the FMN could oxidize external electron donors instead of the natural counterpart Cys, when it is mutated to Ala or Gly. The UV-vis absorption and fluorescence spectroscopy revealed that the photo-activation of the C/A mutant yielded FMNH· with concomitant oxidation of external chemicals at a greater amount compared to the C/G mutant. The results suggested that the suitable mutations including those at the Cys site can provide a possible photo-catalysts to mediate electron transfer between photo-inactive chemical species.

2D1515 Nonlabens dokdonesis DSW-6 由来微生物型ロドプシンの発現
Expression of microbial rhodopsins in Nonlabens dokdonesis DSW-6

Yuichi Hashimoto, Rei Abe-Yoshizumi, Hideki Kandori (*Nagoya Inst. Tech.*)

Recently, a light-driven outward Na^+ pump, KR2, was found from Krokinobacter eikastus. Nonlabens dokdonesis DSW-6 similar marine bacterium as K. eikastus, encodes rhodopsins, outward H^+ and Na^+ pumps. In this study, we examined their pump activity in native cells. When N. dokdonesis DSW-6 is cultivated in MB medium, it showed H^+ pump activity in all growth phase. On the other hand, Na^+ pump activity was detected only in the stage between growth and stationary phases, when it is cultivated under high NaCl concentration. Physiological role of microbial rhodopsins in N. dokdonesis DSW-6 will be discussed.

2D1530 光駆動 Na^+-H^+KR2 におけるイオン取込み機構の研究
Ion uptake mechanism of a light-driven Na^+-H^+ pump, KR2

Yoshitaka Kato[1], Keiichi Inoue[1,2], Rei Abe-Yoshizumi[1], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*PRESTO, JST*)

We recently found a Na^+ pumping rhodopsin from marine bacterium Krokinobacter eikastus (KR2). KR2 pumps Na^+ in NaCl solution, but is converted to H^+ pump in KCl or salts of larger cations [1]. Recent crystal structure of KR2 showed the presence of the intracellular cavity [2], whose role in ion uptake is not clear.
In this study, we measured transient absorption changes under several conditions in order to investigate Na^+- and H^+- uptake mechanism. Our results show that [H^+] or [NaCl] affect the kinetics of blue-shifted M intermediate, which can be well explained by competitive ion-uptake model. Ion selectivity of Na^+ / H^+ will be discussed.
[1] Inoue et al. *Nat Commun.*, 2013, 4, 1678
[2] Kato et al. *Nature*, 2015, 521, 48-53

2D1545 In situ 光照射固体 NMR によるバクテリオロドプシン Y185F 変異体の光反応経路の解明
Photo-reaction pathways of bacteriorhodopsin Y185F mutant as revealed by in situ photo-irradiation solid-state NMR spectroscopy

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Photo-reaction pathways of a bacteriorhodopsin Y185F mutant were examined using in situ photo-irradiation solid-state NMR spectroscopy. The 13-cis, 15-syn (CS; bR548) changed to a CS* (13-cis, 15-syn) at -40 C, and the all-trans (AT; bR568) transformed to an N under green (520 nm) light. In the dark, N transformed to an O at -40 C. Consequently, the O transformed to the N through the AT, whereas the CS* did not change under 520 nm light. The CS* was converted to the AT at -20 C. The AT was converted to the CS at 20 C. The configuration of the AT, CS, CS*, N, and O were determined as (13-trans, 15-anti), (13-cis, 15-syn), (13-cis, 15-syn), (13-cis, 15-anti), and (13-trans, 15-anti), respectively, by inspecting ^{13}C chemical shifts of [20-^{13}C, 14-^{13}C]Ret-Y185F-bR.

2D1600 プロトンドナーを持たない新たなプロトンポンプ型ロドプシン
A new proton pump rhodopsin without a conserved proton donor

Yuto Suzuki[1], Keiichi Inoue[1,2], Leonid S. Brown[3], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*PRESTO JST*, [3]*University of Guelph*)

Microorganisms use light energy for life activities. Bacteriorhodopsin (BR) is the most famous light-driven proton pump. In BR, a proton is transferred to the Asp85 from the retinal Schiff base by light, followed by the Schiff base reprotonation by Asp96. PaR is a proton pump rhodopsin from Pantoea ananatis, a plant pathogen. While proton acceptor(Asp83) is conserved, corresponding position to Asp96 in BR is Gly in PaR. Despite of the lack of proton donor, we found that PaR effectively pumps protons. Here, we study reaction dynamics and structure of PaR in detail by spectroscopic methods, based on which molecular mechanism will be discussed.

2E1355 天然変性タンパク質 HIV-1 Tat の pH 依存的な構造変化
pH-dependent conformational changes in the intrinsically disordered HIV-1 Tat protein

Tomoko Kunihara, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

HIV-1 transactivator of transcription (Tat) is an intrinsically disordered protein, and it binds to zinc ions under physiological pH. Previous studies have focused on the structure of Tat at acidic pH, but the structure of the zinc-bound Tat at physiological pH remains unclear. Here, we studied pH-dependent conformational changes of Tat by CD, NMR, and X-ray scattering. We revealed that Tat is unfolded at acidic pH, and the zinc-free Tat at neutral pH has structures similar to the acid-unfolded state. Moreover, we found that the zinc-bound Tat at neutral pH has more compact structures with residual helices. These results suggest that the extent of disorder in the structures of intrinsically disordered Tat is highly sensitive to pH and zinc ions.

2E1410 野生型及び病原性変異体 β2 ミクログロブリンの圧力変性反応の研究
Insights into the transition state of pressure-induced denaturation of β2-microglobulin and its pathogenic variants

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β2-Microglobulin (β2m) is the causative protein of dialysis-related amyloidosis. It was reported that β2m unfolds along the pathway of N_C -$>I_C$ -$>U_C$ -> U_T, whereas it refolds along the U_T -> I_T -> I_C -> N_C pathway, where the I_T state is a putative amyloidogenic precursor state.
Two aggregation-prone variants of β2m, ΔN6 and D76N, have been so far reported. We observed the equilibrium and kinetics transitions of pressure-induced folding/unfolding on the wild type and the variants. The analysis of the results revealed that the transition from the unfolded state to the putative amyloidogenic state needs a transient volumetric expansion in the wild type, whereas D76N does not need such a large expansion for the transition, making its occurrence easier.

2E1425 ライン共焦点検顕微鏡を用いたマイクロ秒分解一分子 FRET 測定よる高速タンパク質折り畳みダイナミクスの追跡
Tracking microsecond single-molecule FRET dynamics on the fast protein folding by the line-confocal microscopy

Hiroyuki Oikawa[1], Munehito Arai[2], Atsuhito Fukasawa[3,4], Hiroaki Yokota[4], Toru Ide[5], Satoshi Takahashi[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Dept. Life Sci., Univ. Tokyo*, [3]*Hamamatsu Photonics K.K.*, [4]*GPI*, [5]*Grad. Sch. Nat. Sci. and Tech., Okayama Univ.*)

Single-molecule detection of fluorescence resonance energy transfer (sm-FRET) is a powerful technique for the investigation of protein dynamics. We recently introduced the line-confocal system for the sm-FRET measurements and achieved the time resolution of 20 μs; however, the observation time was limited to ~ 1 ms. In this investigation, we newly introduced hybrid photo detectors to our system and obtained the time resolution of 10 μs and the observation time of more than 10 ms. We investigated the equilibrium folding transitions of the B domain of protein A (BdpA) doubly labeled with fluorophores. The traces exhibited the submillisecond dynamics in the unfolded state and the folding transitions.

2E1440 一分子蛍光分光法によって観測されたユビキチンの変性状態の不均一性
Conformational heterogeneity of denatured ubiquitin detected by single molecule fluorescence spectroscopy

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Single-molecule (sm) spectroscopy is useful to elucidate properties of individual proteins. We investigated the folding of ubiquitin at the single-molecule level using line confocal microscope. We obtained time series of sm-FRET efficiency from ubiquitin labeled with two different dyes. At low concentration of denaturant, we obtained traces having higher FRET efficiency corresponding to the native state. At higher concentration of denaturant, the traces possess lower FRET efficiency corresponding to the denatured state. Contrary to the previous concept of the denatured proteins, which fluctuate randomly without any structures, our data suggested that the denatured ubiquitin are heterogeneous and that the components possess the lifetime longer than several milliseconds.

2E1455 多数ペプチドから成る系の全原子分子動力学シミュレーションによるペプチド溶解性
Peptide solubility estimated by all-atom molecular dynamics simulation of multi-peptide systems concur with experimental values

Yutaka Kuroda[1,2], Yuji Sato[2], Atsushi Suenaga[3], Makoto Taiji[4] ([1]*TUAT, Dept Biotech & Life Sci*, [2]*TUAT, Dept Biotech & Life Sci*, [3]*Nihon University*, [4]*Quantitative Biology Center, RIKEN*)

In order to investigate amino acid's contribution to protein and peptide solubility, we carried out 100 ns molecular dynamics simulations of 106 $Å^3$ cubic boxes containing ~$3x10^4$ water molecules and 27 tetra-peptides composed of a single amino acid type. The calculations were performed using AMBER-8 with standard force field on a special purpose MD-GRAPE3 computer, and without introducing any “artificial” hydrophobic interactions. Tetra-peptides composed of I, V, L, M, N, Q, F, W, Y, and H formed large aggregates, those containing A, P, S, and T formed smaller ones, and D, E, K, and R did not aggregate. Overall, this very first all-atom molecular dynamics simulation of multi-peptide systems appears to reproduce the basic properties of peptide solubility.

2E1515 出芽酵母を利用した SOD1 オリゴマー形成メカニズムの解明
A mechanism describing pathological oligomerization of SOD1 in a budding yeast model

Kyohei Onose, Yuko Nishiura, Yoshiaki Furukawa (*Dept. Chem., Keio Univ.*)

Mutations in Cu,Zn-superoxide dismutase (SOD1) cause a familial form of amyotrophic lateral sclerosis (ALS). The misfolded oligomers of mutant SOD1 is abnormally accumulated in affected spinal motor neurons not in the other proliferative cells. It, however, remains obscure why SOD1 misfolding occurs only in the non-proliferative motor neurons. Here, we have noted that budding yeast can be either in a proliferating or non-proliferative state and found that the oligomerization of mutant SOD1 occurs only in a non-proliferating state. Accumulation of oxidative stress in a non-proliferating state is further considered to trigger the formation of abnormal SOD1 oligomers cross-linked via disulfide bonds and thereby decrease the enzymatic activity of SOD1.

2E1530 肝吸虫由来の銅シャペロンに着目した新たな SOD1 活性化メカニズムの解明

A new mechanism of SOD1 activation regulated by copper chaperone in Clonorchis sinensis

Mami Fukuoka[1], Isao Nagano[2], Yoshiaki Furukawa[1] ([1]*Dept. Chem., Keio Univ.*, [2]*Gifu Univ. Sch. Med.*)

Copper chaperone for superoxide dismutase (CCS) is a metallochaperone that delivers a catalytic copper ion to Cu,Zn-superoxide dismutase (SOD1). CCS is composed of three domains, among which a precise role of domain I in the activation of SOD1 remains unknown. Here, we have noted the absence of domain I in Clonorchis sinensis CCS (CsCCS) and found an important role of domain I in the copper transfer to SOD1. CsCCS was not able to activate SOD1 but introduce the disulfide bond in SOD1 that is essential to enzymatic activity. C. sinensis is parasitic on bile duct where copper ions exist abundantly; therefore, C. sinensis SOD1 could acquire a copper ion in a CsCCS-independent fashion, and CsCCS is supposed to function as a sulfhydryl oxidase not a copper chaperone.

2E1545 ペプチドを利用したミスフォールド型 SOD1 タンパク質の新たな検出手法

A new peptide-based method to detect misfolded SOD1 proteins

Kenichi Nagasawa, Takao Nomura, Yoshiaki Furukawa (*Dept. Chem., Keio Univ.*)

Mutations in Cu,Zn-superoxide dismutase (SOD1) cause a familial form of amyotrophic lateral sclerosis. Presence of misfolded SOD1 in the affected motorneurons is proposed as a pathological hallmark of this disease, but it remains quite obscure how much of mutant SOD1 become misfolded under pathological conditions. Here, we have developed a quantitative detection method of misfolded SOD1 by using a peptide that can recognize abnormal conformations of SOD1 with sub-micromolar affinity. More specifically, we have screened the peptide library by a phage display method and successfully identified several peptides that can exclusively recognize misfolded SOD1. Using those peptides with gel-filtration assay, quantitative analysis of misfolded SOD1 was performed.

2E1600 スーパーオキシドディスムターゼにおける新たな金属イオン獲得経路

A new intramolecular route to acquire metal ions in superoxide dismutase

Teppei Kokubo, Yasuyuki Sakurai, Yoshiaki Furukawa (*Dept. Chem., Keio Univ.*)

In metalloproteins, the binding of metal ions is essential to the expression of their enzymatic activities as well as the maintenance of structural stabilities. Cu,Zn-superoxide dismutase (SOD) is a classical metalloenzyme that binds copper and zinc ions, but it still remains obscure how SOD recognizes their metal ions and binds those at their canonical binding sites in vivo. Here we have found that metal ions are first captured by the conserved Cys residues in E. coli SOD and then transferred to their canonical binding sites. Following the transfer of metal ions, furthermore, the Cys residues in SOD were oxidized to form the disulfide bond. We thus propose a new metal binding site in SOD that plays important roles in acquiring metal ions in vivo.

2J1355 超解像光学顕微鏡で観察した、収縮環におけるアクチンフィラメントとミオシンの配置

Arrangement of actin filaments and myosins in contractile ring, revealed with super-resolution microscopy

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Super-resolution microscopes reveal fine structures smaller than resolution limits of conventional optical microscopy. The super-resolution microscope opened the new window to observe biological events of 50-100 nm level.
We were, therefore, interested in obervation of cytokinesis. Many authors reported the presence of filamentous actin and myosin II in the contractile ring but it still remains. unknown how the actin filament and myosin II are arranged to generate force for division.
Here we observed the contractile ring with SIM and STED. We found arrangement of the actin filaments and and myosin II. Mechanism of movement of the ring will be discussed in the presentation.

2J1410 走化性タンパク質の発現量が大腸菌の走化性シグナル伝達系に与える影響

Effect of the expression level of the chemotaxis proteins to the kinetic property of the signal processing of *Escherichia coli*

Takashi Sagawa[1], Hiroto Tanaka[1], Yoshiyuki Sowa[2], Ikuro Kawagishi[2], Hiroaki Kojima[1] ([1]*NICT*, [2]*Dept. of Frontier Biosci., Hosei Univ.*)

Using chemotaxis signal processing system, *Escherichia coli* regulates the rotational direction of their flagellar motors in response to recognition of chemoeffectors. To investigate kinetic property of the signal processing induced by the chemoeffectors in detail, we comprehensively measured time course of directional biases (CW bias) of rotating tethered cells of which expression levels of chemotaxis proteins were regulated and quantified. After the addition of chemoeffectors, change in the CW bias of rotating flagellar motor was delayed about 50-140 ms. And the length of the delay was decreased with a slope of the temporal change in the CW bias. We would like to discuss the cause of the delay and kinetic property of the signal processing in this annual meeting.

2J1425 滑走するバクテリアの自己集合により形成される巨大渦の一方向性回転

Directional rotation of large-scale vortex made of self-assembly of gliding bacteria

Showko Odaka, Daisuke Nakane, Takayuki Nishizaka (*Dept. Phys., Gakushuin Univ.*)

Flavobacterium johnsoniae exhibit gliding motility driven by left-handed helical flow of surface protein. We have reported, in the 52nd meeting, their extraordinary behavior in high density on an agar substrate: multiple vortices with the size of a few millimeter spontaneously appeared. We here found that each vortex was a thin, circular plate made of self-assembly of the bacteria. All plates rotated in a CCW manner at a speed of 0.5 μm/s. To understand the mechanism of this collective motion, the motility of the individual bacterium was analyzed. At low density, each bacterium formed a cluster after collision, and its speed increased 6 times faster. These clusters were subsequently assembled and connected with unknown secretion products to construct a large plate.

2J1440 ゲル上での細胞集合体形成における力学モデルの検討

Mechanical Study of Formation of Cell Aggregates on Hydrogel

Yuko Shimokawa[1], Hiroyuki Koike[3], Takahisa Matsuzaki[1], Masahiro Enomura[2], Masaki Kimura[2], Seiichiro Nakabayashi[1], Hiromitsu Nakauchi[3], Hideki Taniguchi[2], Takanori Takebe[2,3], Hiroshi Yoshikawa[1] ([1]*Dept. Chem., Saitama Univ.*, [2]*Dept. Regen. Med., YCU*, [3]*ISCBRM, Dept. Med., Stanford Univ.*)

Organs develop from cell aggregates, termed 'organ bud', emerging at the early stage of organogenesis. Recently, Takebe and Yoshikawa et al reported that such cell aggregates could be autonomously formed on gel substrates with proper stiffness [1]. It was also revealed that the formation of cell aggregates is mainly driven by myosin-II-based cytoskeletal contractile forces of mesenchymal stem cells, which could be influenced by stiffness (Young's modulus: E) of underlying hydrogel substrate. In this presentation, we explain mechanical model of the cell aggregate formation and its control by using spatial stiffness patterns of gel substrate.

[1] T. Takebe and H. Y. Yoshikawa et al., Cell Stem Cell 16, 556-565 (2015).

2J1455 マウス気管繊毛の非対称運動メカニズム ー軸糸の変形の視点からー

Mechanism of asymmetric beating of mouse tracheal cilia - *from the perspective of the axoneme bending* -

Takanobu A Katoh[1], Koji Ikegami[2], Nariya Uchida[3], Toshihito Iwase[4], Tomoko Masaike[4,5], Mitsutoshi Setou[2], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin Univ.*, [2]*Dept. Cell Biol. and Anat., Hamamatsu Univ. Sch. Med.*, [3]*Dept. Phys., Tohoku Univ.*, [4]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [5]*PRESTO, JST*)

Highly coordinated dyneins generate ciliary asymmetric beating. We herein aim to understand the correlation between the forces and bending deformation of a cilium by the followings: 3-D tracking; optical tweezers; DIC adjusted to the different focal plane. A bead was attached to the tip of the cilium, and trapped at various points of beating stroke to measure the maximum force (F_M) and the stiffness of a cilium. F_M increased from 10 to 80 pN as the point was displaced from the position of post-effective stroke, suggesting that the change in the force is partially attributable to the bending deformation of a cilium. We also obtained the shape of a cilium to analyze the shape by numerical calculation and elucidate the dynein-coordinated mechanism through the deformation.

2J1515 チトクローム c 酸化酵素における酸素還元反応とプロトンポンプ共役機構

The coupling mechanisms of oxygen reduction reaction and proton pumping in cytochrome c oxidase

Satoru Nakashima, Tatsuhito Nishiguchi, Chen Li, Kyoko Shinzawa-Itoh, Shinya Yoshikawa, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

The coupling mechanisms of oxygen reduction reaction and proton pumping of cytochrome c oxidase have been studied by using time-resolved vibrational and absorption spectroscopy. From the observed results, multi parts of the protein cooperate each other to attain its proton pumping function. At the first stage of the reaction, the ligand oxygen attached to CuB before binding to heme a3. Since the oxygen affinity of heme a3 seems to correlate with hydration state of water channel of H-pathway, its role is monitoring the timing of the start of the reaction. Accompanied with this ligand binding to heme a3, α-helix structure (bulge structure) along proton pump pathway, changed its orientation to close the gate of the water channel, which protects the back leak of protons.

2J1530 蛍光減少率測定によるヘムオキシゲナーゼ変異体ー電子供与蛋白相互作用の解析

Fluorescence decay rates reveal the unique interactions between heme oxygenase mutants and its electron-donor protein

Norio Miyake, Taiko Migita (*Fac. of Agri., Dep. of Biol. Chem., Yamaguchi Univ.*)

Heme degradation by HO requires electrons and molecular oxygen. The electron is supplied from NADPH carried by a protein unique to each bio-species: cytochrome P450 reductase (CPR) for animals and ferredoxin reductase-ferredoxin (FNR-Fd) for plants as well as cyanobacteria. Our research, thus far, on HOs from different biological species strongly suggests the importance of the electron-transfer efficiency in the HO activity. In this study, we focused on the natural fluorescence from HO or Fd and from FMN or FAD in CPR or FNR. The fluorescence from CPR/Fd-FNR varied its intensity by the addition of heme-HO complexes. The fluorescence-decay rate was compared among heme-HO mutants of rat and soybean, which gave new insights into the inter-protein interactions.

2J1545 亜硝酸還元酵素と一酸化窒素還元酵素の複合体形成による効率的な一酸化窒素分解機構

Formation of Complex between Nitrite Reductase and Nitric Oxide Reductase for Rapid NO Elimination

Takehiko Tosha[1], Erina Terasaka[1,2], Kimi Matsumoto[1,2], Hiroshi Sugimoto[1], Yoshitsugu Shiro[1,2] ([1]*RIKEN SPring-8*, [2]*Grad. Sch. Sci., Univ. Hyogo*)

Denitrification is a form of anaerobic respiration, which produces N2 from nitrate via four reduction steps. While, in this process, cytotoxic nitric oxide (NO) is produced as an intermediate product, denitrifying bacteria can survive. This indicates that denitrifying bacteria have a system to suppress the diffusion of NO. Here, we found a formation of the complex of NO-forming nitrite reductase (NiR) and NO reductase (NOR) as a possible system for effective NO elimination. The structure of the NiR-NOR complex was solved at a resolution of 3.2 Å, but no obvious pathways for NO channeling from NiR to NOR was observed. We, therefore, propose that NiR produces NO at the close proximity to NOR by the complex formation, facilitating NO binding to NOR for rapid NO reduction.

2J1600 マイクロ流路フローフラッシュ赤外吸収分光法の開発と一酸化窒素還元酵素の反応中間体解析への応用

Development of Micro-channel Flow-flash Infrared Absorption Spectroscopy and its Application to Intermediate of Nitric Oxide Reductase

Tetsunari Kimura[1], Shoko Ishii[1,2], Takehiko Tosha[1], Yoshitsugu Shiro[1,2], Minoru Kubo[1,3] ([1]*SPring-8 Cent., RIKEN*, [2]*Grad. Sch. Sci., Univ. Hyogo*, [3]*PRESTO, JST*)

Characterization of the transient intermediates in the chemical and biological reactions is essential to clarify their molecular mechanisms. In this research, we have developed an infrared spectroscopic system by combining laser spectroscopy and micro-channel flow-cell, which enabled us to monitor the irreversible enzymatic reaction of low-yield membrane protein with the nanosecond time resolution. We applied this system to the catalytic reaction of nitric oxide reductase (NOR). NOR is a membrane protein, which reduces NO to N_2O at the reactive center consisting of heme *b* and non-heme Fe_B. The reaction was initiated by the NO diffusion from caged NO, and the changes in the N-O stretching vibrational modes were observed. The molecular mechanism of NOR will be discussed.

2K1355 タンパク質フォールディングの動的機構の理論的研究
Theoretical study of the dynamic protein folding/unfolding transition mechanisms

Toshifumi Mori, Shinji Saito (*IMS*)

The understanding of protein folding has greatly advanced over the last few decades. Computational and experimental tools can follow μ~ms dynamics in atomistic details, and are used to study the transition mechanisms in a synergistic manner. Yet, interpretation of the dynamic mechanisms remain somewhat controversial, partly due to the high-dimensional nature of the system.
Here we study two fast-folding proteins by analyzing the ultralong trajectories from D. Shaw et al. We find that the transition dynamics is heterogeneous, and involves multiple transition paths with intermediate and misfolded states. This implies the need to understand the mechanism beyond a one-dimensional picture. We further discuss how the heterogeneity may appear in experimental observables.

2K1410 Mechanisms of selection of channel for substrate transport in GatCAB

Jiyoung Kang, Masaru Tateno (*Grad. Sch. Sci., Univ. Hyogo*)

In translational systems of eubacteria, glutamine amidotransferase CAB (GatCAB) converts Glu-tRNAGln to Gln-tRNAGln (because of the absence of eubacterial GlnRS). In GatCAB, two catalytic sites, i.e., the glutaminase and transamidase sites, are utilized to produce ammonia species and transfer of ammonia to the tRNA, respectively. However, the two sites are ~30 Å apart, and so ammonia is transported from the former to the latter in the enzyme. In our previous molecular dynamics simulations, we revealed that among two possible pathways, a hydrophobic channel exhibits a significant preference for ammonia transport. In this study, we further analyzed the detailed mechanisms that bring about the differences of the preferences, by comparing the two pathways/channels.

2K1425 Characterizing the gating of Skp: a periplasmic chaperone

Daniel Holdbrook[1], Bjorn Burmann[2], Sebastian Hiller[2], Peter Bond[1] ([1]*Bioinformatics Institute, A*STAR*, [2]*Biozentrum, University of Basel, Basel, Switzerland*)

In this molecular dynamics (MD) simulation study, we demonstrate the dynamic nature of the outer membrane protein-binding compartment of the Skp chaperone, and estimate the upper limit of cargo sizes it can accommodate. Previously, using a combined NMR spectroscopy and MD approach, we showed that the release of Skp's cargo is unlikely to be triggered by a unique lipid, lipopolysaccharide, found in the bacterial outer membrane, due to the absence of a specific binding site. In the current study, we observe a conformational change in Skp, and rotation about a putative hinge region, resulting from stored elastic strain between two helices that form the boundary of the binding compartment. This motion may reflect the ability of Skp to adapt to substrates of different sizes.

2K1440 トレオニン合成酵素における生成物支援触媒機構についての理論的解明
Theoretical elucidation on the molecular mechanism of product assisted catalysis of threonine synthase

Mitsuo Shoji[1], Yuzuru Ujiie[1], Megumi Kayanuma[1], Yasuteru Shigeta[1], Takeshi Murakawa[2], Hideyuki Hayashi[2] ([1]*University of Tsukuba*, [2]*Osaka Medical College*)

Threonine synthase (TS) catalyzes the last step of L-Thr biosynthesis and its reaction is the most complex among the PLP enzymes. To elucidate the detailed mechanism, we performed comparative Quantum Mechanics/ Molecular Mechanics calculations with an exhaustive search for the reaction pathways in the reaction-specificity-determining-process. Satisfactory agreements with the experimental data were obtained. Contrary to the earlier proposal, the base that abstracts a proton from the attacking water was the Lys61 amino group rather than the phosphate ion. We also determined that phosphate ion forms a stable H-bond with the L-Thr moiety, which is critical for the reaction specificity of the product assisted catalysis.

2K1455 Reaction mechanism of glycinamide ribonucleotide synthetase: Free-energy profile of the formation of an acylphosphate intermediate

Norifumi Yamamoto[1], Gen-ichi Sampei[2], Gota Kawai[1] ([1]*Chiba Tech*, [2]*Univ Electro Comm*)

Glycinamide ribonucleotide (GAR) synthetase called PurD catalyses the second reaction of the de novo purine nucleotide biosynthetic pathway, yielding GAR from phosphoribosylamine (PRA), glycine and ATP. The mechanism of the GAR synthetic reaction in PurD consists of two partial reactions and, in this presentation, we report a theoretical perspective of the molecular mechanism of the first half, where glycine reacts with ATP to yield an acylphosphate intermediate. We revealed a free-energy profile along the minimum energy path (MEP) of the reaction with QM/MM simulations combined with an MEP search algorithm and a free-energy perturbation method. The energetics charting the free-energy profile helps in the comprehensive understanding of the reaction process.

2K1515 Exploring N-glycan conformers: assessment of force fields and enhanced sampling algorithms

Raimondas Galvelis, Suyong Re, Yuji Sugita (*RIKEN TMSL*)

N-linked oligosacharides (N-glycans) play an important role in protein folding control, cell adhesion, organism immune response, etc. They functional versatility come from structural variety and the inherit flexibility of glycosidic bonds, which results into many distinct conformers. However, it makes experimental characterisation and computational modelling challenging. In this work, we assess the accuracy of established force fields and enhanced sampling algorithms (REMD and RSE-MTD, our improved metadynamics scheme) on a biologically relevant N-glycan (Bi9). In particular, we focus on the estimation of conformer populations, which are compared with the experimental NMR measurements.

2K1530 Oligomerization of Aβ fragments by the Hamiltonian replica-permutation method

Satoru Itoh[1,2], Hisashi Okumura[1,2] ([1]*IMS*, [2]*Sokendai*)

The amyloid-β peptides (Aβ) form amyloid fibrils which are associated with Alzheimer's disease. It is necessary to clarify the oligomerization process of Aβ in order to understand the amyloid fibril formation process and to find a remedy for Alzheimer's disease. We applied the Hamiltonian replica-permutation method (HRPM) to Aβ fragments to study their oligomerization process. HRPM combines the advantages of RPM and the Hamiltonian replica-exchange method (HREM). RPM is a better alternative to REM. In RPM temperature permutations among more than two replicas are performed with the Suwa-Todo algorithm. In HREM, by exchanging the parameters that are related only to limited degrees of freedom, the number of replicas can be decreased in comparison with REM.

2K1545 拡張アンサンブル分子動力学シミュレーションを用いた電子顕微鏡データからの構造精密化手法
New Approach for Cryo-EM Data Flexible Fitting Using Generalized Ensemble Molecular Dynamics Simulation

Osamu Miyashita[1], Chigusa Kobayashi[1], Takaharu Mori[1,2], Yuji Sugita[1,2], Florence Tama[1,3] ([1]*RIKEN AICS*, [2]*RIKEN TMSL*, [3]*Nagoya Univ. Phys.*)

Cryo-EM single particle analysis provides structural information on biologically important molecular complexes. However resolution is still not sufficiently high, and thus, high-resolution structures from other experiments are often used to create an atomic model from such low-resolution structural information.
An approach for building such atomic models is the use of molecular dynamics simulation with biasing forces that guides the model to fit into the low-resolution cryo-EM data. However it can fail to construct models for complex systems. We present a new approach that employs replica-exchange algorithm to enhance the sampling of conformations and suitable biasing force parameters, which improves the accuracy of the atomic models derived from Cryo-EM data.

2K1600 単原子イオン周囲のハイパーモバイル水は多体効果に起因する
Hypermobile Water around Monoatomic Ions Is Derived from Many-Body Effect

George Mogami[1], Nobuyuki Matubayasi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Eng. Sci., Osaka Univ.*)

Many of ions were found to have hypermobile water (HMW), which had higher dielectric relaxation (DR) frequency than bulk water (G. Mogami et al., 2013). Since HMW is thought to affect interactions among proteins or polymers, it is necessary to understand a nature of HMW at a molecular level to evaluate thermodynamic properties of proteins or polymers including hydration state. In this study, thermodynamic quantities of ionic solvation were calculated and separated into ion-water (two-body) and water-water (many-body) interactions on the basis of the molecular dynamics and energy representation method (N. Matubayasi et al., 2008), resulting in a beautiful correlation between the DR and thermodynamic values and a major contribution of many-body term to HMW formation.

2L1355 Novel algorithm for identification of gene clusters in whole genome DNA sequences by combining rigorous and heuristic schemes

Yuhya Takahashi, Jiyoung Kang, Masaru Tateno (*Grad. Sch. Life Sci., Univ. Hyogo*)

It was believed the gene order was randomly accomplished in eukaryotic genomes; however, recent studies reported that some functionally-related genes are clustered with the very short intergenic distances in eukaryotic genomes. The two properties of the clusters (i.e., functional relationship and physical distance) are, in principle, common to those of operon in eubacterial genomes. In this study, we developed a novel algorithm to identify gene clusters in whole genome DNA sequences. It was revealed that the obtained clusters were closely correlated to the active regions specified as the chromatin loop domains that were experimentally identified by three-dimensional mapping techniques between the distinct genome sites.

2L1410 DELTA-FORTE: a profile-profile comparison method enhanced by curated database

Toshiyuki Oda[1], Kazunori Yamada[1,2], Kentaro Tomii[1] ([1]*Biotech. Res. Inst. for Drug Discovery, AIST*, [2]*Grad. Sch. Info. Sci., Tohoku Univ.*)

Sequence similarity search and alignment methods are indispensable in modern biology. There is still room for improvement, though many algorithms and tools have been developed. We have proposed a profile-profile alignment method, called FORTE (FOld Recognition TEchnique), which is suitable for identifying a distant relationship and/or structural similarity between proteins. This time, we present an upgraded version of FORTE, DELTA-FORTE by using DELTA-BLAST and CDD for its profile construction. This implementation gave us improvements in terms of both runtime and sensitivity. In benchmarking with the SCOP20 dataset, the accuracy of DELTA-FORTE outperformed proven existing methods, such as PSI-BLAST, HHsearch, and a previous version of FORTE.

2L1425 A Clustering Approach to Visualize the Sequence-Structure-Function Relationship of Protein Enzymes

Te-Lun Mai, Geng-Ming Hu, Chi-Ming Chen (*Department of Physics, National Taiwan Normal University*)

Protein enzymes play essential roles in various biological and pathological processes, and therefore many drugs are designed to target these enzymes. The purpose of this work is to provide an integrated clustering approach, using minimum span clustering and minimum spanning tree algorithms, to examine and visualize the sequence-structure-function relationship of proteins. From our clustering results, we discuss possible divergent evolution and convergent evolution of protein enzymes, and a close sequence-structure-function relationship was observed for them. Our results validate this integrated approach in clustering the structural fold and function class for protein enzymes, and support the prediction of protein function using sequence information alone.

2L1440 ホモ二量体タンパク質の立体構造変化の構造的、機能的特徴
Structural and functional characterization of structural changes in homodimeric proteins

Takayuki Amemiya, Tatsuya Horii, Ryotaro Koike, Motonori Ota (*Grad. Schl. of Info. Sci., Nagoya Univ.*)

Most of proteins form protein complexes to function, and their structures change in many cases when functioning. We examined structural changes in homodimeric proteins extensively, as the first step to study structural changes in protein complexes. The structural changes of 2831 homodimeric proteins were collected from PDB and they were classified into 6 motions. Among them, the 'over' and the 'interface' motions showed significant differences in some properties at an interface between two protomers. In addition, we analyzed the correlations between molecular functions and the motions using GO terms. The results of classification, interface analyses and functional characterization will be presented in detail.

2L1455 ハブタンパク質のいるところ
Where the hub proteins are

Motonori Ota[1], Hideki Gonjya[1], Ryotaro Koike[1], Satoshi Fukuchi[2] ([1]*Grad. Sch. Info. Sci., Nagoya U.*, [2]*Fac. Eng., M.I.T.*)

Protein-protein interaction is fundamental for all biological phenomena, and whole view of the interactions is represented by the protein-protein interaction network. The hub proteins having a lot of interaction partners play the vital role in the network. We investigated the sub-cellular locations of proteins in the network, and found that the proteins localized in the multi sub-cellular components, especially the nucleus and cytoplasm, tend to be hub proteins. Examination on keywords suggested that many of them change states and transfer by the post-translational modifications, and function for transcription. The other characters are discussed in terms of the multi-domain proteins and intrinsic disorder.

2M1355 細いフィラメント上のトロポミオシンの位置を ESR 距離測定により探索する
Searching for tropomyosin position in the thin filament by distance measurements using spin-labeling dipolar EPR spectroscopy

Toshiaki Arata[1], Keisuke Ueda[2], Yoshiki Tsujimoto[1], Masao Miki[3] ([1]*Dept. Biol., Sci. Grad. Sch., Osaka Univ.*, [2]*RIKEN-Yokohama*, [3]*Univ. Fukui*)

The mobility of a spin label on tropomyoin (Tm) side chain showed unexpectedly no Ca2+ effect. Here, distance distribution was obtained by fitting the dipolar interacting spectrum of 15N-based spin labeled Tm and 14N-based spin labeled Cys374 of actin after non-interacting spins of actin Cys374 was cancelled. Without Ca2+ the distance were found to be 2-2.5 nm only at the N- and C-terminal regions of Tm. Distance between spin labeled Tm and actin (Mn2+) were also determined to be 2-2.5 nm at some Tm residues but beyond sensitivity (>2.5 nm) at the other over a wide region of Tm. We are constructing the model of Tm positioning on actin filament by 2-nm distance search of Tm residues from Mn2+ and Cys374 of actin.

2M1410 骨格筋ミオシン分子間の力発生が同調する仕組み
Mechanism of cooperative force generation between skeletal myosins

Motoshi Kaya[1], Takumi Washio[2], Toshiaki Hisada[2], Hideo Higuchi[1] ([1]*Graduate School of Science, University of Tokyo*, [2]*Graduate School of Frontier Science, University of Tokyo*)

Skeletal myosins are packed into a filamentous array so that they can interact simultaneously with a single actin filament. Thus, we simply ask the following question; "can the force generation between skeletal myosins be synchronized?" To address the question, we measured the force generated by synthetic myofilaments by using optical tweezers. The stepwise force generations were consistently observed against high loads beyond 30 pN at 1, 0.1, and 0.01mM ATP respectively, implying that individual steps were likely driven by synchronous activities between myosins. Combined with a two-working stroke simulation model, we proposed that synchronous force generations between myosins could be more frequently provoked by higher loading and ATP conditions.

2M1425 蛋白質ダイナミクスから観た筋収縮調節機構：筋肉の細いフィラメントの中性子散乱による研究
A view of the regulatory mechanism of muscle contraction from protein dynamics: a neutron scattering study of muscle thin filaments

Satoru Fujiwara[1], Tatsuhito Matsuo[1], Takeshi Yamada[2], Kaoru Shibata[3] ([1]*QuBS, JAEA*, [2]*CROSS-Tokai*, [3]*J-PARC Center*)

In order to investigate the regulatory mechanism of muscle contraction in terms of protein dynamics, we carried out neutron scattering experiments on the native thin filaments (NTF) in the presence and absence of Ca^{2+} and F-actin using the dynamics analysis spectrometer *DNA* at J-PARC. The elastic incoherent and quasielastic neutron scattering experiments showed that NTF in the -Ca^{2+} state is more flexible than in the +Ca^{2+} state, and that this difference in flexibility arises from the different distributions of the local atomic motions. Comparison with F-actin suggests that the differences arise from the regulatory proteins. These results imply that regulation of the protein dynamics plays an important role in the regulatory mechanism of muscle contraction.

2M1440 心筋の昇温誘起高速サルコメア振動
Hyperthermal Sarcomeric Oscillations in cardiac muscle

Seine Shintani[1,2], Hideo Higuchi[1], Norio Fukuda[3], Shin'ichi Ishiwata[4] ([1]*Dept. Phys., Univ. Tokyo*, [2]*JSPS Research Fellow*, [3]*Dept. Cell Phys., Sch. Med., Univ. Jikei*, [4]*Dept. Phys., Univ. Waseda*)

Sarcomere is an essential unit of cardiomyocyte. It is well known that sarcomere has three state (i.e. contraction, relaxation and spontaneous oscillation state) dependent of the solution condition. In intact cardiomyocytes, we found hyperthermal sarcomeric oscillations (HSOs) that is Ca^{2+} independent and high-frequency (~5-10 Hz) auto-oscillations induced by a rapid increase in temperature to >~38 ° C. This finding suggest that the temperature shift the state from relaxation to oscillation. We found that the HSOs was occurred in skinned cardiac myofibrils. The skinned myofibrils is suitable for exchanging chemical compounds and proteins. Therefore using this system, we will understand the molecular aspect of HSOs.

2M1455 **Disorder profile of nebulin encodes a vernier-like position sensor**

Ming-Chya Wu[1,2], Jeffrey G. Forbes[3], Kuan Wang[2,4] ([1]*RCADA, NCU, Taiwan*, [2]*IP, AS, Taiwan*, [3]*TWIC, Inc, USA*, [4]*IBC, AS, Taiwan*)

Nebulin is long intrinsically disordered scaffold for the thin filaments of skeletal muscle sarcomere. It wraps around actin filament, stabilizes thin filaments and regulates Ca-dependent actomyosin interactions. We address whether the disorder profile of nebulin might encode guidelines for thick and thin filament interactions, by analyzing the disorder probability of human nebulin which correlates with structural features of the thin filaments. We identify hidden periodicities in both the nebulin disorder profile and the A band stripes in the electron micrographs of the sarcomere. The cross-correlation analysis at various sarcomere length depicts a Vernier-like design for both periodicities, thus enabling nebulin to sense position and fine tune sarcomere overlap.

2M1515 **電気刺激により引き起こされるグラスキャットフィッシュの電気受容求心性線維の発火の非線形特性**

Nonlinear characteristics of electrosensory afferent nerve impulses elicited by sinusoidal electric stimulation in glass catfish

Yu Adachi, Katsumi Tateno (*Kyushu Institute of Technology*)

We studied nonlinear responses to sinusoidal electric stimulation in afferent nerves of ampullary electroreceptors of glass catfish (Kryptopterus bicirrhis/minor). Spontaneous afferent nerve impulses were modulated by the external sinusoidal electric stimulation (1-100 Hz) via the ampullary electroreceptors. The one-dimensional return map revealed nonlinear responses of the afferent nerve impulses. The one-dimensional return map showed a set of line segments or a triangular closed line in the frequency range between 2 and 40 Hz. In those 1D maps, a stable fixed point was not found. Those results indicate that the sinusoidal electric stimulation induced non-periodic responses of the afferent nerve impulses.

2M1530 **膜電位感受性色素による神経回路機能のアッセイ系の構築ー海馬スライスとビスフェノール A**

Neural circuit functional assay with voltage-sensitive dye imaging in hippocampal slices; effect of maternal bisphenol A

Takashi Tominaga[1], Yoko Tominaga[1], Katsuhide Igarashi[2,3], Maky Otsuka I[2,3], Yusuke Furukawa[2], Jun Kanno[2], Kentaro Tanemura[4] ([1]*Inst. Neurosci., Tokushima Bunri Univ.*, [2]*Div Cellular & Molecular Toxicol, NIHS*, [3]*L-StaR, Hoshi Univ. Sch. Pharmacy Pharmaceutical Sci.*, [4]*Lab Animal Reproduction, Grad Sch Agr Sci.*)

We aimed to show that voltage-sensitive dye imaging (VSDI) works for brain neural circuit dysfunctions. We examined the effect of maternal exposure to bisphenol A (BPA), which may be a risk of delayed emotional and cognitive deficits in the offspring. BPA-containing drinking water was supplied from mating until weaning and subjected to the experiments after eight weeks. Three main pathways of the hippocampus (HP) were stimulated and examined with VSDI. In the HP slices of control mice, even very weak stimulation was sufficient to induce large and long responses in the entire CA3-CA1, whereas the HP slices of BPA-exposed mice did not exhibit such responses. This difference may indicate an alteration in the threshold of excitability of the cells.

2M1545 **フェムト秒レーザー光刺激による神経細胞ネットワークの機能解析**

Functional analysis of living neuronal networks with a femtosecond laser-induced stimulation

Yuta Nakagawa[1,2], Suguru N. Kudoh[2], Takahisa Taguchi[3], Chie Hosokawa[1] ([1]*Biomed. Res. Inst., AIST*, [2]*Grad. Sch. Sci. & Tech., Kwansei Gakuin Univ.*, [3]*CiNet, NICT*)

To understand information processing in neuronal networks, it is important to identify functional connections between neurons. A precise stimulation of a single neuron induced by a focused femtosecond laser is powerful tool to study spatio-temporal dynamics of the neuronal activity. When a femtosecond laser was focused on a neuron loaded with a fluorescent calcium indicator, fluorescence intensity rapidly increased at the cell body. Subsequently, fluorescence intensity in other neuronal cell close to the target neuron increased, suggesting that the propagation on neuronal activity was induced by the femtosecond laser. Moreover, we analyzed synchrony of the neuronal activity after femtosecond laser stimulation with raster plots, which will be presented and discussed.

2N1355 **補酵素置換による微生物外膜シトクロムの電子伝達速度制御**

Rate Regulation of *in vivo* Extracellular Electron Transport by Replacing Flavin Cofactors in Outer-membrane *C*-type Cytochromes

Yoshihide Tokunou, Akihiro Okamoto, Kazuhito Hashimoto (*Department of applied chemistry, Univ. Tokyo*)

Metal reducing bacteria localize *c*-type cytochromes at the outer membrane (OM *c*-Cyts), which enables electron transfer from the cell inside to the outside. We recently found that non-covalent flavin cofactor in OM *c*-Cyts likely take an essential role for electron and proton transport across OM. Here, we found that replacing flavin cofactor with analogous molecules with different redox potential and pK_a could control the rate of electron transport via OM *c*-Cyts *in vivo*. This finding provides not only the clue for cofactor recognition in OM *c*-Cyts, but also the opportunity for designing optimal cofactor.

2N1410 **Uniquely Small Outer Membrane Cytochrome-c as a Possible Electron Carrier for Direct Electron Uptake in a Sulfate Reducing Bacterium**

Xiao Deng, Akihiro Okamoto, Kazuhito Hashimoto (*Grad. Sch. Eng., Univ. Tokyo*)

Sulfate reducing bacteria (SRB) has been thought to enhance the rate of anaerobic corrosion by oxidizing organics or H2 to produce corrosive H2S. Meanwhile, specific SRB such as Desulfovibrio ferrophilus IS5 (strain IS5), are recently proposed to conduct direct electron extraction from Fe0, causing intense iron corrosion. In this study, we found that abundant cytochrome-c (13 kDa) localized in the outer membrane which likely plays an essential role in the microbial electron extraction mechanism. Its molecular size is uniquely smaller than other reported outer membrane electron transfer enzymes. We will discuss structural features of the outer membrane enzyme based on our genome sequence analysis of strain IS5.

2N1425 酸素センサータンパク質 Aer のシグナル伝達機構の解明
Elucidation of signal transduction mechanism of Aer

Yoriyoshi Oka, Tatsuya Iwata, Masayo Iwaki, Hideki Kandori (*Nagoya Institute of Technology*)

Aer is a membrane protein for aerotaxis in E. coli. Aer has FAD-bound PAS domain as a sensor domain. It is proposed that the redox reactions of FAD cause structural changes of PAS, followed by activation of signaling domain.
In order to reveal the signal transduction mechanism of Aer, we investigated the spectroscopic properties by UV-vis and FTIR spectroscopy. First, we constructed expression system of Aer in E. coli and optimized purification conditions. Next, we investigated reducing reagents that reduce FAD of Aer, and we found that sodium dithionite can reduce FAD. We then obtained difference FTIR spectra originating from reduction/oxidation of FAD. Structured changes of Aer and signaling mechanism will be discussed based on these spectroscopic results.

2N1440 一分子 FRET 観察によるセンサリーロドプシン-トランスデューサー複合体の構造変化の研究
Single-molecule FRET studies on the light-induced structural changes of the sensory rhodopsin-transducer signaling complex

Ryo Nishimura[1], Keiichi Inoue[1,2], Jin Yagasaki[3], Kenichi Kawamoto[1], Yuki Sudo[4], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*JST PRESTO*, [3]*Nagoya University*, [4]*Okayama Uiversity*)

Sensory rhodopsin II (SRII), is a photo-receptor membrane protein having vitamin-A aldehyde retinal as a chromophore. SRII is responsible for negative phototaxis in a collaborative work with its cognate transducer protein, HtrII, in membrane. To observe conformational changes and structural fluctuations in the complex upon illumination, here we employed single-molecule fluorescence-resonance-energy-transfer (FRET) method.
In this study, we constructed two cycteine mutants, V185C for SRII and D102C for HtrII, and they were purified and labeled with dyes independently. Based on the FRET signal from the receptor-transducer complex, light-induced structural changes will be discussed.

2N1455 銅輸送 P 型 ATPase における ATP 加水分解中の イオン結合構造の ESR 解析
ESR Spectroscopy on Metal binding Sites in Cu^{2+}-Transporting Protein during ATP Hydrolysis

Satoshi Yasuda[1], Takuya Horimoto[1], Hiroaki Daimon[1], Yasuhiro Ueda[1], Naoyuki Kuwabara[3], Toshiaki Arata[1,2] ([1]*Dept. Biol. Sci., Osaka Univ.*, [2]*Ctr. High Mag, Field Sci., Grad. Sch. Sci., Osaka Univ.*, [3]*Photon Factory, KEK*)

Using ESR spectroscopy, we saw a paramagnetic ion being transported inside pump ATPase. The spectra from T. thermus CopB showed the presence of 2 classes of Type-2 Cu^{2+} center at high (2 moles) and low (~8 moles) affinity in which Cu^{2+} are coordinated in a distorted square pyramidal geometry with nitrogen ligands. About 3 moles of Cu^{2+} ions are bound to a CopBΔN ATPase molecule in which the N-terminal 30 residues are truncated, indicating that N-domain contained ~6 Cu^{2+} sites as a reservoir. We found slight or no difference after ATP or AMPPCP addition, suggesting that weakly bound coordination of Cu^{2+} was formed only transiently during transport. Distances between Cu^{2+} and spin label and between labels will be measured to see transport pathway and domain movements.

2N1515 ES 細胞の分化における単一ヌクレオソームのイメージング
Single nucleosome imaging in the differentiation of embryonic stem cells

Mai Tambo[1,2], Tadasu Nozaki[1,3], Sachiko Tamura[1,2], Kazuhiro Maeshima[1,2] ([1]*Natl. Inst. of Genet.*, [2]*Sch. of Life Sci. The Grad. Univ. for Adv. Studies*, [3]*Inst. for Adv. Biosci., Keio Univ.*)

Human genomic DNA wraps around histone proteins to form the 10-nm nucleosome fiber. Recent studies including our researches suggested that nucleosome fibers are folded irregularly in nucleus without regular and higher order chromatin fiber. This irregular folding implies a physically less restricted and more dynamic state. To confirm this dynamic chromatin state, we established a single nucleosome imaging system and found that nucleosomes fluctuated locally in living cells. To investigate the difference of chromatin dynamics between differentiated and undifferentiated cells, we observed nucleosome fluctuation in mouse embryonic stem cells. We would like to discuss the change of chromatin dynamics through cellular differentiation and its meaning.

2N1530 幹細胞の確率的分化ダイナミクスのつくる空間パターン
Spatial Pattern in Stochastic Dynamics of Stem Cell Differentiation

Hiroki Yamaguchi[1], Kyogo Kawaguchi[2], Takahiro Sagawa[3] ([1]*Dept. Bas. Sci., Univ. Tokyo*, [2]*Dept. Sys. Biol., Harvard Med. Sch.*, [3]*Dept. Appl. Phys., Univ. Tokyo*)

Adult mammalian tissue is typically maintained by stem cells. Much attention has been paid to the stochastic nature of the differentiation dynamics of stem cells. Two different theoretical scenarios have been suggested, in order to explain the experimentally observed macroscopic statistical behaviors. Especially in 2D tissues, these two scenarios give the same statistical law. Thus the differentiation dynamics is not enough to determine the genuine scenario in the differentiation dynamics of stem cell. In this talk, we describe the spatial dynamics of stem cells as well as the differentiation dynamics, and discuss the spatial pattern of the dividing stem cells which has never been focused previously, in order to investigate which scenario would be valid in vivo.

2N1545 成長する上皮管の径サイズを維持する動的な細胞挙動について
On Dynamic Cellular Behaviors Realizing Stable Radial Size in Developing Epithelial Tubule

Tsuyoshi Hirashima, Taiji Adachi (*Inst Front Med Sci, Kyoto Univ*)

A diameter of epithelial tubule in the developing murine epididymis is almost constant in time and space during the elongation; there should exist some mechanisms at the cell level for maintaining the tubule diameter. However, it remains unclear what cellular behaviors control the radial size of developing tubule. We identified actomyosin-based cellular constriction at the apical junctions of epithelial tubule is required to maintain the diameter, and found a remarkable cellular behavior driven by the actomyosin constriction. Furthermore, a multicellular dynamic model suggested that such a cellular behavior would explain the homeostatic tubule radial size. We propose a possible mechanism of cellular behavior in controlling the radial size of developing tubule.

2N1600 枯葉に擬態した蝶の模様の進化
Evolution of leaf mimicry in butterfly wing patterns

Takao K Suzuki, Shuichiro Tomita, Hideki Sezutsu (*National Institute of Agrobiological Sciences*)

How leaf mimicry evolved has fascinated many biologists, but it remains unclear. Here we show evolutionary origin and process of leaf wing patterns of butterflies (*Kallima*). We used comparative morphological analyses and Bayesian phylogenetic methods to estimate the past wing patterns and the temporal order of character state changes, and revealed that the leaf pattern has evolved through several intermediate patterns from a non-mimetic ancestor. Our study provides the first evidence for gradual evolution of leaf mimicry. Furthermore, we investigated the difference between evolutionary process of leaf mimicry and that of lichen cryptic patterns of butterflies (*Hamadryas*), which reveal that leaf patterns evolved in a contingent fashion.

2O1355 PELDOR study on the high-affinity Mn(II) site of photoactivation of photosystem II

Mizue Asada, Hiroyuki Mino (*Grad. Sch. Sci., Nagoya Univ.*)

Photosynthetic oxygen evolution is carried out by Mn cluster in Photosystem (PS) II. Mn cluster consists of four Mn, one Ca and five O ions and surrounding amino acids. Assembly of the Mn cluster to PS II is a light-induced process, which is called as 'photoactivation'. The process of photoactivation starts with incorporation of one Mn(II) to a high-affinity site in apo-PS II in dark state. It has not clarified where the high-affinity site for Mn(II) is located within the oxygen evolving complex. We measured the distance between the redox-active tyrosine residue Y_D and the Mn(II) binding to the affinity site by Pulsed electron-electron double resonance (PELDOR). The PELDOR signals indicated that the Mn(II) locates at the position of Mn4.

2O1410 時間分解 EPR 法によるホウレン草の光合成光化学系 II 反応中心における初期電荷分離構造
Time-Resolved EPR Study on Geometry and Dynamics of the Primary Charge-Separated State in the Photosystem II Reaction Center of Spinach

Yasuhiro Kobori[1], Masashi Hasegawa[1], Takashi Tachikawa[1], Toru Kondo[2], Hiroki Nagashima[3], Takahiro Sakai[3], Hiroyuki Mino[3] ([1]*Graduate School of Science, Kobe University*, [2]*Tokyo Institute of Technology*, [3]*Graduate School of Science, Nagoya University*)

In the initial stage of the photosynthesis of the plants, photoinduced sequential electron-transfers are known to take place in the photosystem II (PSII) reaction center. It has been suggested that the photoinduced charge-separated (CS) state composed of P(D1) radical cation and pheophytin radical anion is generated via the excited singlet state of the accessory chlorophyll. However, no experimental studies have been performed to determine the geometries of the primary CS state in the protein. We have analyzed the time-resolved EPR spectra of the excited triplet state to characterize the geometries and the exchange coupling of the primary CS state in D1D2cytb559 core of PSII.

2O1425 FTIR spectroelectrochemical measurement of the redox potential of the secondary quinone electron acceptor Q_B in photosystem II

Yuki Kato, Ryo Nagao, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

Photosystem II (PSII) drives electron transfer (ET) from the Mn cluster to two quinones, Q_A and Q_B. The driving force of the ET between these quinones is their redox potential (E_m) gap. However, the E_m gap value is still unclear, because E_m of Q_B has not been directly measured. Meanwhile, $E_m(Q_A)$ has been estimated to be ca. –100 mV in intact PSII, and found to shift by ~+150 mV by inactivation of the Mn cluster. In this work, we measured $E_m(Q_B)$ by applying FTIR spectroelectrochemistry to reveal the E_m gap. $E_m(Q_B)$ was determined to be +159 mV in intact PSII, and shifted by –26 mV by Mn depletion. We thus conclude that Mn depletion decreases the E_m gap from ~260 to ~80 mV and hence promotes the reverse ET, leading to photoprotection when the Mn cluster is inactivated.

2O1440 光化学系 II におけるチロシン Y_D からのプロトン放出：FTIR 法による検出
FTIR detection of proton release from the redox-active tyrosine Y_D in photosystem II

Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

Photosystem II has two redox-active tyrosines, Y_Z and Y_D. Although these tyrosines are symmetrically located, they have significantly different functions. Whereas Y_Z is a major electron donor of P680, Y_D functions only as a peripheral electron donor. To clarify the origin of this difference, we studied where the proton from Y_D is transferred to upon its oxidation using FTIR spectroscopy. Isotope-edited signals of Mes buffer in a $Y_D^{\cdot}/Y_D$ difference spectrum showed that about 0.84 protons were released into the bulk upon oxidation. It is thus concluded that the proton of Y_D is released out of the proteins, whereas Y_Z transfers its proton to a neighboring His through a hydrogen bond. This long distance proton transfer was suggested to cause the slow redox reactions of Y_D.

2O1455 光合成水分解反応における Yz 周辺の水素結合ネットワークの役割
FTIR study on the role of hydrogen bond network around Yz during photosynthetic water oxidation

Ryo Nagao, Hanayo Ueoka-Nakanishi, Takumi Noguchi (*Grad. Sch. Sci., Univ. Nagoya*)

The X-ray crystal structure of photosystem II revealed two major proton transfer pathways: a hydrogen bond network around Cl ion and that via the redox-active tyrosine Yz. To examine the role of the Yz network during water oxidation, we analyzed a site-directed mutant of D1-N298, which interacts with D1-H190 hydrogen-bonded with Yz. Fourier transform infrared analysis showed that the D1-N298A mutant decreases the efficiency of the S2 to S3 transition and blocks the S3 to S0 transition. Moreover, structural changes in a hydrogen bond network and in the interactions of a weakly hydrogen-bonded water molecule(s) were observed. These results suggest that the hydrogen bond network via Yz functions as a proton transfer pathway during the S2 to S3 and S3 to S0 transitions.

2O1515 DNA の結晶構造中の構造ゆらぎの解析
Reconsidered DNA conformations in crystal structures

Tomoko Sunami, Hidetoshi Kono (*JAEA*)

Conformational flexibility of DNA plays important roles on biological processes such as transcriptional regulations. Therefore, it is important to analyze when and how DNA shows conformational variations.
Electron density maps in crystallographic analyses contain information of conformational variations. But such information is typically discarded in the refinement process. To extract the information about conformational variations, we have performed comprehensive analysis of the electron density maps of DNA crystals.
We found that positive FoFc densities are observed more frequently around phosphates than around bases, indicating high flexibility of DNA phosphates. In the presentation, we will discuss more details.

2O1530 Temperature Dependence of Structural Dynamics of RNA and DNA Hairpins Studied by 2D Fluorescence Lifetime Correlation Spectroscopy

Chao-Han Cheng[1], Kunihiko Ishii[1,2], Tahei Tahara[1,2] ([1]*Molecular Spectroscopy Laboratory, RIKEN*, [2]*Ultrafast Spectroscopy Research Team, RIKEN Center for Advanced Photonics*)

In this study, we investigated the temperature-dependent structural dynamics of FRET dye-labeled RNA and DNA hairpins by 2D fluorescence lifetime correlation spectroscopy (FLCS) to elucidate the origins of difference in the opening and closing processes between RNA and DNA. The 2D FLCS revealed a marked difference in the temperature dependence of the closing process between RNA and DNA. The closing rate of RNA is more sensitive to temperature compared to that of DNA which is almost unchanged at various temperatures. It is suggested that the closing process of RNA contains an additional activation enthalpy barrier that arises from the intrachain interaction of loop.

2O1545 DNA 酵素による紫外線損傷 DNA 光修復の構造解析
Structural analysis of DNAzyme that functions as photorepair of UV-damaged DNA

Yuhi Kurahashi, Tatsuya Iwata, Hideki Kandori (*Nagoya Inst. Tech*)

DNAzyme is a DNA that shows enzymatic activity. Although there is no report of DNAzymes discovered in vivo, DNAzymes with various enzymatic activities have been created by in vitro selection method. In 2004, DNAzyme that repairs UV-induced damaged DNA using >305 nm light was reported. This DNAzyme, named UV1C, has guanine rich sequence and shows enzymatic activity in the presence of Na+ or K+, so that UV1C is considered to form a higher order structure, G-quadruplex. In order to clarify the repair mechanism of UV1C, we try to measure the DNA repair by FTIR spectroscopy. After confirmation of the repair of TDP substrate by UV1C in the presence of Na+, we applied FTIR spectroscopy to the DNAzyme-substrate complex.

2O1600 クロマチンは塩によってどのようにコンパクトになるのか？
How can chromatin condense with salt? - a model study using a synthetic nucleosome system

Kazuhiro Maeshima[1], Takaaki Hikima[2], Ryan Rogge[3], Jeffrey Hansen[3], Sachiko Tamura[1] ([1]*National Institute of Genetics*, [2]*RIKEN SPring-8 Center*, [3]*Colorado State University*)

How a long strand of genomic DNA is organized in the cell remains as a basic question in cell biology. DNA is wrapped around the core histone octamer to form a nucleosome fiber. It is well known that cations induce nucleosomes into a large structure in vitro. Recently we found that interphase and mitotic chromatin consist of irregularly folded nucleosome fibers without a regular 30-nm chromatin fiber. To examine whether this irregular folding is true for the cation-induced large structure, we conducted a structural study of synthetic 12-mer nucleosome arrays using small-angle x-ray scattering (SAXS) analysis at SPring-8. The obtained results showed that the cation-induced large structure also consists of irregularly folded nucleosome fibers.

2P1355 多様な昆虫の翅外形を記述する力学モデル
A mechanical model for diversified wing margin shapes among insects

Yukitaka Ishimoto (*Fac. of Mach. Int. & Sys. Eng., Akita Pref. U.*)

Insect wings in nature exhibit astonishing morphological diversity. The wings of some insects such as flies develop smooth margin shapes, implying the existence of a highly organized multicellular mechanical structure.
We modeled the margin shape of insect wings as an Euler's elastica. Our simulation showed that the distribution of the bending stiffness of the wing margin could reproduce a diversity of margin shapes. We further estimated the distribution of the bending stiffness from experimental images of D. melanogaster wings (Ishimoto, Sugimura, Morishita, in preparation). We will show the effect of the intrinsic tension of the wing blade, and discuss about an influence from the veins in the end.

2P1410 数理モデルとイメージングを用いた多階層連結による走電性の理解
Integration of Multilayer Stages in ElectroTaxis By Using Live Imageing and Simulation

Masato Yasui[1], Satomi Matsuoka[1], Masahiro Ueda[1,2] ([1]*RIKEN*, [2]*Osaka University*)

Cells sense the gradient of the electric field and migrate to the electrode, which is called "Electro-taxis," and is used in wound healing. Because it is possible to control the electric field quantitatively, electrotaxis is suitable to research how cells sense the field. To understanding the mechanism of taxis, we observed single molecule PTEN and dynamics of phosphatidyl inositol lipid system, which is used in cell migration and gradual sensing. Then, we have constructed the model integrating the results of the multi-layer imaging data. As the result, we have understood PTEN and the phosphatidyl inositol lipid system are used for stabilizing the cell migrating direction to the electrode in high efficiency.

2P1425 増殖集団における細胞状態の重要性を評価する 1 細胞統計手法

Single-cell statistics to evaluate the significance of cellular state in growing population

Takashi Nozoe[1], Yuichi Wakamoto[1,2] ([1]*Grad. Sch. Arts and Sci., Univ. Tokyo*, [2]*Research Center for Complex Systems Biology, Univ. Tokyo*)

Recent advances of single-cell observation techniques have revealed variation and fluctuation of cellular state on a large scale. To understand the biological role of such heterogeneity, it is critical to evaluate cost and benefit of cellular state deviating from population average. We previously developed the statistical method to evaluate the potential for reproduction or “fitness” of state of cell lineage. This approach also provides a measure of “significance" of cellular state. Our experimental data suggests that our measure of significance represents the intrinsic correlation between cellular state and the division frequency, and that in sub-lethal ribosome-targeted antibiotic treatment the transcriptional activity of its resistant gene has great significance.

2P1440 簡単な細胞モデルを用いた Pirt 方程式の導出

Derivation of the Pirt equation in the simple cellular model

Yusuke Himeoka, Kunihiko Kaneko (*Tokyo Univ. Department of Arts and Sciences*)

Pirt equation is important empirical relationship about efficiency of cellular growth. This equation claims the linear relationship between growth speed and nutrient consumption rate of microorganisms. Therefore, it is of great interest to study Pirt equation for both understanding fundamental nutritional usage of cell and application to industry such that cellular engineering.
However, there are no theoretical understandings of the equation. Thus, I tried to derive Pirt equation in microscopic cellular model. In my talk, I will report some achievements in the derivation in the equation.

2P1455 遺伝子発現及びエピジェネティック修飾ダイナミクス間のタイムスケール

The time scale between gene expression and epigenetic modification dynamics

Tadashi Miyamoto, Kunihiko Kaneko (*Grad. Sch. Art. Sci., Univ. Tokyo*)

The interaction between genes consist of gene regulatory network (GRN), which govern the protein expression dynamics. In addition, it is suggested that epigenetic modification solidifies the cellular state transition. Cell differentiation and reprogramming are interpreted as the cellular state transition by these dynamics. However, the inter-relationship between the gene expression and epigenetic modification dynamics is not fully uncovered.
We then constructed GRN model including epigenetic modification. As a result of simulation, we found that our model showed different dynamics depending on the time scale of epigenetic modification. We are planning to evaluate the consistency between these dynamics and biological phenomenon.

2P1515 ネットワーク構造のデザイン原理と構成要素の応答性

Network Design Principles and Response Sensitivity of Components

Masayo Inoue[1], Kunihiko Kaneko[2] ([1]*molprof, AIST*, [2]*Univ. of Tokyo*)

In this study, we discuss how characteristic features of an evolved network differ according to the response sensitivity of its components. We investigated the responses of gene regulatory networks containing many genes that have undergone numerical evolution. Although we studied with same conditions except for gene sensitivity, the evolved networks were totally different depending on the sensitivity. Networks with sensitive genes evolved with shortest path structures. On the other hand, networks with non-sensitive genes evolved with cooperative structures indecomposable to small motifs. We also studied the robustness of the two types and found cooperative networks were more robust. These results have significant implications in designing robust biological networks.

2P1530 触媒反応ネットワークにおける資源不足と多様化

Diversification by limitation of multiple resources in a catalytic reaction network

Atsushi Kamimura, Kunihiko Kaneko (*Dept. of Basic Science, The University of Tokyo*)

Cells integrate diverse molecules to keep their reproduction taking essential resources from environment. By considering a protocell model consisting of mutually catalyzing molecules and respective resources, we show limitations in the resources result in reproduction of diverse compositions of protocells. As abundances of each resource component are limited, transition from protocells of few components to that with coexistence of diverse components occurs. The number of coexisting species increases with a negative power of the resource abundances. This diversity scaling is estimated from the optimum growth speed of a protocell, as is determined by a tradeoff between utility of diverse resources and concentration onto fewer components to increase the reaction rate.

2P1545 全成分タンパク質合成反応モデルの構築とこれを用いた反応ダイナミクス解析

Reaction dynamics analysis of the whole protein translation system by computational modeling

Tomoaki Matsuura[1], Yoshihiro Shimizu[2], Kazufumi Hosoda[3], Naoki Tanimura[4], Tetsuya Yomo[5] ([1]*Department of Biotechnology, Osaka University*, [2]*Qbic, RIKEN*, [3]*Institute for Academic Initiatives, Osaka University*, [4]*Mizuho Information and Research Institute*, [5]*Department of Bioinformatic Engineering*)

The protein synthesis using the in vitro protein translation system (IVT) involves the reactions with wide range of rates; however, the system reaches a steady-state in less than a minutes. To reveal how such large-scale reaction network can rapidly reach the steady-state, computational model was constructed based on the whole components of the reconstituted IVT that consist only of essential components. The constructed large-scale kinetic model showed similarity with the experiments: the rate of the synthesized peptide reached constant in less than a minutes, and the rate was in a same range as the experiment. We also found that time development of the protein synthesis involves a collapse and a regrowth of the reaction network before reaching the final steady-state.

2P1600 Polynomial-life model towards analysis of turnover and regeneration

Hiroshi Yoshida (*Kyushu Univ. Dept. Math.*)

Most of life maintains itself through turnover, namely cell proliferation, movement, and elimination. Inspired by such a fact, and together with various operations of polynomials, I here propose *polynomial-life* model towards effective analysis of cell turnover and regeneration. Polynomial-life is multicells that are expressed as multivariable-polynomials. A cell is expressed as a term of polynomial, in which point (m,n) is described as a term $x^m y^n$ and the condition is as its coefficient. Further cell elimination, proliferation and movement are expressed as various operations of polynomials. In this framework I shall present various patterns through the polynomial-life model and discuss patterns maintained through turnover.

2Q1425 Molecular Gate Locations in MthK Potassium Channels

Crina Nimigean (*WCMC*)

Ion channel gating is fundamental to cellular signaling. Gates that control K+ channels were found both at a bundle crossing and at the selectivity filter, but the specific location of the gate that opens Ca2+-activated K+ channels has remained elusive. Using the MthK prokaryotic homolog and a stopped-flow fluorometric assay for fast channel activation, we show that intracellular quaternary ammonium blockers bind to closed channels suggesting that the gate must be at the selectivity filter. The blockers access the closed slower than the open channel, indicating that the intracellular entryway narrows upon gate closure, without preventing K+ access. Thus, Ca2+-dependent gating in MthK occurs at the selectivity filter with coupled movement of the intracellular helices.

2Q1355 植物の傷害応答性・長距離・高速カルシウムシグナル伝達

若手招待講演

Mechanical wounding/insect attack-induced, long-distance, rapid calcium signal transduction in plants

Masatsugu Toyota[1,2], Simon Gilroy[1] ([1]*University of Wisconsin-Madison*, [2]*JST, PRESTO*)

Plants sense local stresses (i.e., insect attack) and transmit signals throughout their bodies to acquire resistance against present/future stresses. However, the molecular basis for such rapid, plant-wide signaling remains largely unknown. Using Arabidopsis plants expressing GFP-based Ca^{2+} indicators, we visualized the mechanical wounding/insect attack-induced spatial and temporal dynamics of cytosolic Ca^{2+} in real time. A wound-induced Ca^{2+} increase propagates through the phloem to distal target leaves at a speed of 1,000 μm/s and this movement requires plant glutamate receptor channels. Thus, plants have a unique cell-to-cell and long-range communication system using Ca^{2+}/glutamate signaling networks and trigger systemic resistance responses.

2Q1440 全反射赤外分光で見る電位依存性プロトンチャネル VSOP への金属結合

Metal binding to the voltage-gated proton channel VSOP studied by ATR-FTIR

Masayo Iwaki[1,2], Kohei Takeshita[3,4,5], Yasushi Okamura[2,6], Atsushi Nakagawa[2,3], Hideki Kandori[1,2] ([1]*Nagoya Inst. Tech.*, [2]*JST-CREST*, [3]*Inst. Protein Res., Osaka Univ.*, [4]*Inst. Acad. Initiat., Osaka Univ.*, [5]*JST-PRESTO*, [6]*Grad. Sch. Med., Osaka Univ.*)

The voltage-gated proton channel (VSOP) has a Zn^{2+}-binding site in the extracellular region of voltage-sensing transmembrane helices. Zn^{2+}-induced difference ATR-FTIR spectra of VSOP showed IR features that can be assigned to the His-CN and carboxylate-OCO^- stretches, which are typical to heavy metal chelation, as well as amide I changes likely in α-helical peptide bonds. Based on the IR data, the Zn^{2+}-effects on the protein conformational changes will be discussed in relation with the recent obtained X-ray crystal structure [1] of the Zn^{2+}-bound resting form of VSOP. ([1], Takeshita et al. (2014) Nat. Struc. Mol. Biol., 21, 352.)

2Q1410 パターン化モデル生体膜を用いた、光シグナル伝達に関わる膜タンパク質の脂質ラフト親和性解析

Raftophilicity of membrane proteins in the phototransduction cascade evaluated in a patterned model membrane

Yasushi Tanimoto[1], Sakiko Kojima[1], Kenichi Morigaki[1,2], Fumio Hayashi[3] ([1]*Graduate School of Agricultural Science, Kobe University*, [2]*Organization of Advanced Science and Technology Research Center for Environmental Genomics, Kobe Univ.*, [3]*Graduate School of Science, Kobe University*)

Signal transduction in the phototransduction cascade is believed to be regulated by lipid rafts. We evaluated the affinity of some of the membrane proteins involved in the cascade (rhodopsin (Rh), transducin (Gt), and S-modulin) to lipid raft (raftophilicity) by using a model membrane having patterned liquid ordered (Lo) (raft model) and liquid disordered (Ld) (non-raft model) bilayer domains. We reconstituted the membrane proteins into the model membrane and quantified their partitioning in the Ld and Lo domains. The proteins studied showed preference to the Ld domains with varied degrees. We discuss on the obtained raftophilicities and their implications to the functional roles of lipid rafts.

2Q1455 ナトリウムポンプロドプシンの電気生理学

Electrophysiology of Na^+ pumping rhodopsins

Satoshi Tsunoda[1,2], Hideki Kandori[1,2] ([1]*Nagoya Institute of Technology*, [2]*OptoBioTechnology Research Center*)

The microbial rhodopsins are a class of membrane proteins with seven transmembrane helices harboring an all-trans-retinal retinal chromophore. Recently the first Na^+-pumping rhodopsins (NaRs) were discovered from several marine flavobacteria. When heterologously expressed in *E.coli*, they transport Na^+ under physiological conditions, and transport H^+ in the absence of Na^+. It is important to know expression levels of these rhodpsins in mammalian cells, because relatively high expression is required for electrophysiolgical studies and for optogenetics application of these rhodopsins. We expressed more than 10 already known Na^+-pumping rhodopsins in mammalian cells and studied the pumping functions by a conventional electrophysiology.

1Pos001 電子顕微鏡を用いた核ラミンの動的構造変化
Observation of dynamics of nuclear lamin using electron microscopy

Muneyo Mio[1], Mai Tsunoda[1], Hayato Yamashita[2], Toshihiko Sugiki[3], **Kazuhiro Mio**[1] ([1]*AIST*, [2]*Grad. Sch. Eng. Sci., Univ. Oosaka*, [3]*IPR, Univ. Oosaka*)

The nuclear lamins are type V intermediate filament proteins that constitute lamina at the inner nuclear membrane. Mutations in A type lamins cause various diseases such as muscular dystrophy, lipodystrophy, and progeria syndrome. The center of rod domain of lamin A, there is a flexible discontinuity region called "L12". Observation of AFM shows that L12 aggressively flexible in solution. Using other methods, we confirmed that L12 is important for self-polymerization. Some lamin A related diseases may associate with this abnormality of polymerizetion. Using affinity grid technique, we constructed artificial nuclear lamina-like structure. This mesh-formed lamina mimic can be used for analysis of onset mechanisms of laminopathies.

1Pos002 Single-particle analysis of *Thermus thermophilus* V-ATPase using an electron microscopy

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The V-ATPase from *Thermus thermophilus* is the rotary enzyme that mediates the coupling between ATP synthesis/hydrolysis in V_1 and proton translocation across membranes through V_o. Atomic resolution structural information about the soluble catalytic region is not obtained yet, and little is known about the V_o. Thus, how protons pass through the membrane has not been described in detail. Structural information about the proton translocation region is required.
We have been trying to obtain the structural information about the V-ATPase using electron cryo-microscopy equipped with a direct electron detector. In this presentation, we will report the progress of the single particle analysis of both the V_1 and the intact V-ATPase.

1Pos003 分子動力学法と分子ドッキング法を用いたエストロゲンレセプターのリガンド結合予測
Prediction of binding pose of estradiol to human estrogen receptor: identification of druggable pocket and ensemble-based docking

Hiroaki Saito, Hidemi Nagao, Kazutomo Kawaguchi (*Kanazawa University*)

We developed a new ensemble-based docking method, which is combined the docking method with molecular dynamics (MD) simulations to predict an accurate ligand-receptor binding pose. We applied this method to a prediction of the binding pose of estradiol (E2) to the human estrogen receptor (ER). The predicted binding pose of E2 by the ensemble-based docking method was in good agreement with that of crystal structure of the ER. Further analysis revealed that the score values improve when the Q value of the receptor decreases; the best binding pose was found in the opening event of the binding pocket.

1Pos004 クライオ電子顕微鏡を用いた B 型肝炎ウイルスの表面抗原粒子の構造解析
Structural analysis of hepatitis B surface antigen particles with cryo-electron microscopy

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Sera from the patients infected with hepatitis B virus (HBV) contain the double-layered mature infectious particles known as Dane particles and small noninfectious particles (22-nm diameter) consisted of HB surface (HBs) antigen. HBs particles are much more abundantly observed than Dane particles, and are important as a marker for HBV infection. HBV has four genotypes with different epitopes. However, their structural difference is not fully understood. In order to reveal their detailed structure and to support efficient production of vaccines, we focused on two major subtypes in Japan, adr and adw. The HBs particles were purified from the infected blood, and studied their structure using cryo-electron microscope (EM).

1Pos005 NLRP3 タンパク質リガンド結合ドメインの構造特性
Structure characterization of the ligand-binding domain of an inflammation-related protein NLRP3

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NLRP3 is a proinflammatory protein which is activated by various ligands associated with cellular stress. To investigate molecular details of its activation, we have produced a recombinant leucine-rich repeat domain of NLRP3 (NLRP3-LRR), an N-terminal region considered to play a role as a receptor. We constructed an *E. coli* expression system in which NLRP3-LRR was produced as a His tagged protein. As a result, a purified protein was successfully obtained, although it was prone to aggregate. Along with efforts of optimizing solution conditions to prevent aggregation, a series of characterization of NLRP3-LRR was performed and structural properties associated with the receptor activity will be discussed on the basis of the obtained results.

1Pos006 コレラ菌走化性受容体のリガンド認識機構
Ligand recognition mechanism of chemoreceptor proteins of *Vibrio cholerae*

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Vibrio cholerae, the etiological agent of cholera, is a Gram-negative bacterium with a single polar flagellum. *V. cholerae* has at least 45 genes for Methyl-accepting chemotaxis protein (MCP)-like proteins (MLPs). Among them Mlp24 is required for production of cholera toxin upon mouse infection and bind multiple amino acids. Mlp37, the closest homolog of Mlp24, is known to bind taurine in addition to multiple amino acids. We have solved the structures of the ligand binding domain of Mlp24, its complex with Arg, Asn or Pro and that of Mlp37 in complex with Arg, Ser or taurine. The structures indicated that ligand binding induces a large conformational change of the PAS-like domain and a gap of the entrance of the binding pocket allows the binding of multiple ligands.

1Pos007 EMDB と PDB データの形状類似検索: Omokage 検索
Shape similarity search for EMDB and PDB: Omokage search

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Hybrid methods (combination of X-ray, NMR, EM, SAXS, MD, etc.) elucidate increasing number of structure data of large and complicated assemblies, which are stored in PDB and EMDB. It is often difficult to search these from the database by existing search services. We have developed a web-based service, Omokage search (URL: http://pdbj.org/omokage/) to find data having similar molecular shapes from ~200,000 structure data in EMDB and PDB. It uses both feature-vector and superposition comparisons. First, the feature-vector comparison roughly extracts the similar data from the database. Next, the detected data are examined in detail by the superimposition in 3D space using Gaussian mixture model, it clearly shows similar and different parts of two maps or models.

1Pos008 Porphyromonas gingivalis の T9SS によって分泌される PGN_0123 の結晶化
Crystallization of PGN_0123 secreted by T9SS of Porphyromonas gingivalis

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Porphyromonas gingivalis, the gram-negative anaerobic bacterium, is a major periodontal pathogen and causes tooth loss in industrial nations. The virulence factors, such as gingipain, are secreted through the type 9 secretion system (T9SS) to the cellular surface, and are fixed outside of the cell by sugar chain modification. The proteins secreted by T9SS have a characteristic domain at C-terminal region called CTD. Among them, PGN_0123 affects sugar chain modification of other virulence proteins secreted by T9SS. To elucidate the molecular mechanism of T9SS secretion and the sugar modification, we expressed, purified and crystallized PGN_0123. The crystals were grown from the solution containing PEG10K as a precipitant. The structure analysis is now in progress.

1Pos009 レジストリーの異なるダイニンストーク領域の二次構造解析
Secondary structure analysis of the entire stalk region with two different registries

Haruka Iwasaki[1], Yosuke Nishikawa[2], Momoko Inatomi[1], Hideaki Tanaka[2], Genji Kurisu[2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Institute for Protein Research*)

Motile activity of cytoskeletal motor proteins requires precise coupling between the ATPase and track-binding activities. In dynein, the stalk region separates these two activities with a long coiled coil, so that it must facilitate communication between them. This communication is mediated by a small amount of helix sliding in the coiled-coil. However, there is less structural characterization of the state transition between α and +β registries using the same construct. We performed SDS-PAGE and CD spectra measurements of cytoplasmic and axonemal dynein stalks locked in either registry. We will discuss the secondary structure analysis of two dynein stalks in parallel with an improvement of previously reported X-ray structure of the entire stalk.

1Pos010 水溶性セレノキシド試薬を用いた α-lactalbumin の酸化的フォールディング
The oxidative folding of α-lactalbumin under mild basic conditions by using a selenoxide reagent

Reina Shinozaki, Michio Iwaoka (*Tokai University*)

α-Lactalbumin (αLA), which has four native disulfide bonds (SS), is stabilized by a calcium ion (Ca2+) bound in the β-domain. It was reported that the oxidative refolding from the fully reduced form (R) to the native state (N) proceeds through characteristic intermediates, 2S* and 3S*, at pH 8.4 which are metastable intermediates among the ensembles of the SS intermediates. In this study, to reinvestigate the oxidative folding pathways, we carried out the experiments by application of a selenium oxidant, DL-trans-3,4-dihydroxy-1-selenolane oxide (DHSox), which has high oxidizing ability in a wide applicable pH range (pH 3-10). As a result, the oxidative folding pathways of αLA previously proposed were reproduced.

1Pos011 分子動力学シミュレーションを用いた Hras-GTP/GDP 複合体周辺の水の動きの解析
Analysis of dynamics of water molecules near the Hras-GTP/GDP complexes by molecular dynamics simulations

Takeshi Miyakawa[1], Ryota Morikawa[1], Masako Takasu[1], Kimikazu Sugimori[2], Kazutomo Kawaguchi[2], Hiroaki Saito[2], Hidemi Nagao[2] ([1]*Tokyo Univ. of Pharm. and Life Sci.*, [2]*Kanazawa Univ.*)

In order to understand the mechanism of hydrolysis of GTP in the Hras-GTP complex, we study the structures of the Hras-GTP/GDP complexes in water solvent by molecular dynamics (MD) simulations.
We evaluated the potential parameters around Mg2+ in Hras-GTP/GDP complexes by quantum chemical calculations. We performed MD simulations of the Hras-GTP/GDP complexes in water solvent using parameters of AMBER03 and our parameters around Mg2+. We found that the positions and orientations of water molecules near GTP are different from those near GDP.
In this study, we analyze the dynamics of the individual water molecules near GTP/GDP in the Hras-GTP/GDP complexes.

1Pos012 プロテイン A とその誘導体の溶液内構造。X 線小角産卵法による研究。
Structure of Protein A and its derivative in solution studied by small-angle x-ray scattering

Masaji Shinjo[5], Kaoru Ishimura[6], Akitsugu Yamamoto[6], Hiroshi Kihara[1,2,3,4] ([1]*Himeji Hinomoto College*, [2]*Hinomoto Gakuen Educational Foundation*, [3]*Ritsumeikan Univ. SR Center*, [4]*Nagoya Univ. SR Research Center*, [5]*Kansai Medical Univ*, [6]*Nagahama Inst of Bio-Sci. & Tech*)

Protein A (PA) is a 56 kDa surface protein with five homologous Ig-binding domains found in the cell wall of S. aureus. Each domain folds into a three-helix bundle, and is able to bind to IgGs. PA releases Igs in acidic pH, suggesting its pH-dep. conformational change. We, then, start investigating the structure of PA and its derivative, C5, in both of neutral and acidic pHs by SAXS. Results show: PA and C5 form globule conformations, and their Rg values are 3.73 ± 0.02 nm within experimental errors both at neutral and acidic pHs. However, Kratky plots and P(r) suggest the conformation of PA at acidic pH is different from the other cases. Experiments were done at PF BL 6A with the approval No. of 2013G101. Authors are grateful to the sample supply from KANEKA CORP.

1Pos013 分子動力学シミュレーションによる Aβ アミロイド線維の揺らぎと構造
Structure and fluctuation of Aβ fibril by molecular dynamics simulations

Hisashi Okumura[1,2], Satoru G. Itoh[1,2] ([1]*IMS*, [2]*SOKENDAI*)

Amyloids are insoluble and misfolded fibrous protein aggregates and associated with more than 20 serious human diseases. To overcome these diseases, it is essential to understand amyloid genesis and disruption. For example, amyloid-β fibrils (Aβ) are known to be associated with the Alzheimer's disease. The structure of Aβ42, consisting of 42 amino acids, has two intermolecular β-sheet regions in the amyloid fibrils. It is known that the Aβ fibril extends only in way, but the reason is still unknown. To answer this question, we performed all-atom molecular dynamics simulations of an amyloid-β fibril in explicit water. We found that the fluctuation and structure are different at both ends of the fibril. This difference may cause the difference in the fibril extension.

1Pos014 tRNA チオ化酵素 TtuA-TtuB 複合体の結晶構造解析
Crystal structure analysis of TtuA-TtuB, a tRNA thiolation enzyme complex

Shun Narai[1], Minghao Chen[1], Naoki Shigi[3], Yoshikazu Tanaka[2], Min Yao[2] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Facl. of Adv. Life Sci., Hokkaido Univ.*, [3]*Biotech. Res. Inst. for Drug Discov., AIST*)

Posttranscriptional modifications are essential for tRNAs to express their functions properly, and more than 90 kinds of modifications are found so far. Among them, sulfur-containing modifications provide various important features to tRNA. The 2-thioribothimidine at the position of 54 (s2T54) enhances structural stability of tRNA, which is introduced by Two thiouridine synthase A (TtuA) and TtuB. Although it is known that TtuA and TtuB act as thiolation enzyme and sulfur-carrier protein, respectively, their detailed sulfur transfer mechanism is still unclear. In the present study, we determined crystal structure of TtuA-TtuB complex. Based on the revealed structure, we discuss the sulfur transfer mechanism by these two proteins.

1Pos015 The approach toward crystallization of HBV core of genotype C

Katsumi Omagari, Yasuhito Tanaka (*Dept. Virol., Grad Sch. Med., Nagoya City Univ.*)

Hepatitis B virus (HBV) is a major human pathogen that causes serious liver disease. HBV has been classified into 8 geographically, genetically, and clinically divese genotypes A to H. Genotype C is prevalent in Japan. The virus consists of a host-derived lipid envelope containing surface proteins, nucleocapsid (HBV core) and a partially double-stranded DNA genome. HBV replicates through reverse transcription of an RNA intermediate, the pregenomic RNA (pgRNA). The replication occurs inside HBV cores. Knowledge of the structure of HBV core would be valuable for understanding the molecular basis. No empirical structural data exist for HBV core of genotype C. This work has established large expression system of HBV core of genotype C toward revealing the crystal structure.

1Pos016 Development of a De Novo protein structure prediction: generating new fold structures by permutating and reversing SSEs of known folds

Shunsuke Nishiyama, Kodai Takagi, George Chikenji (*Grad. Sch. of Engineering, Nagoya Univ*)

Success of De Novo protein structure prediction is limited to small proteins with simple topology. One of the major bottlenecks for predicting structures of larger proteins with complex topology is conformational sampling. To overcome this problem, we developed a new method for generating novel structures. The method permutates Secondary Structure Elements (SSEs) and reverses the N- to C-terminal direction of SSEs of currently existing protein structures, while prohibiting unusual topologies such as crossing loops and left-handed beta-alpha-beta cross over connections. In the presentation, we will provide the detail description of the algorithm and the results of the stucture prediction benchmark test.

1Pos017 コフィリンの結合によるアクチン線維構造変化の解析
Elucidation of structural change in actin filament invoked by cofilin

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An eucaryotic protein cofilin severs the actin filament, and also accelerates the depolymerization rate. In this study, to reveal the structure change in the actin filament invoked by cofilin, we determined the cryo-EM density map of the cofilin decorated actin filament (cofilactin) at ~7Å resolution. As previously reported, cofilactin has different helical symmetry than naked actin filament, owing to the conformational change of actin-subunit into monomeric-like twisted form (Galkin et al., 2011). Further, we observed the conformational transition of DNaseI-binding loop into α-helix like form, the salt-bridge pare switchings in actin-actin interface. We are now investigating the role of them in the cofilin function by biochemical/structural assays of mutant proteins.

1Pos018 MP2/6-311G++(d,p)法による炭酸脱水酵素の機能発現と構造に関する理論的研究
Basis Set Having Diffuse Function, 6-311G++(d,p), in MP2 to Calculate Energy of Active Site Model Containing His/Trp in Carbonic Anhydrase

Muhamad Koyimatu[1], Kimikazu Sugimori[2], Hidemi Nagao[2], Hideto Shimahara[1] ([1]*Japan Advanced Institute of Science and Technology*, [2]*Kanazawa University*)

His64 in the active site of carbonic anhydrase is well-known to have a pH-dependent change between two conformations, "in" and "out". For a long time, it has been discussed whether a rotational or swinging behavior between the "in" and "out" is possible in the catalytic CO_2 hydration reaction or not. Here, the rotational properties in the model structure that has Trp5 and Gly63-His64-Ala65 with/without a water-bridge system including zinc ion were examined by using the basis set, 6-311G++(d,p), combined with three functionals (B3LYP, M06-2X, and MP2). In MP2, we found that the use of 6-311G++(d,p) causes the decrease of the interaction energy values between His64 and Trp5 compared to the use of 6-31G(d,p), whereas such dependency is not seen in B3LYP and M06-2X.

1Pos019 多剤排出トランスポーター AcrB の Motion-Tree 法による解析

Motion tree analysis of the multidrug transporter AcrB

Tsutomu Yamane[1], Ryotaro Koike[2], Motonori Oota[2], Satoshi Murakami[3], Akinori Kidera[1], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City Univ.*, [2]*Grad. Sch. of Inf. Sci., Nagoya Univ.*, [3]*Grad. Sch. of Biosci. and Bioeng., Tokyo Inst. of Tech.*)

AcrB is one of the proton-driven multidrug transporters from the Gram-negative bacteria E. coli., and forms homo trimer with different conformations that are sequentially converted during drug export. These cyclic conformational changes are called functionally rotating mechanism. We have studied about the structural changes through functionally rotating mechanism using the Motion-Tree analysis, which describe the structural change as rigid body motions, and found the key motions to transfer the proton motive force to the drug export pathway. In the present study, we analyzed the water pathways in trans-membrane domain, which are important for proton transfer. We will show the detail of the results in the poster presentation.

1Pos020 シアノバクテリア由来のフィトクロム Cph1 の光受容部位の構造変化ダイナミクス

Conformational dynamics of photosensory domains of the Cyanobacterial Phytochrome 1 (Cph1)

Kimitoshi Takeda, Masahide Terazima (*Grad. Sch. Sci., Kyoto Univ*)

Cyanobacterial phytochrome 1 (Cph1) is a red and far red light sensor protein which reversibly photoconverts between Pr and Pfr states. Cph1 consists of N-terminal photosensory domains and a C-terminal kinase domain. In previous work, it was reported that the construct which contained only sensory domains formed monomer in Pr state and dimer in Pfr state, which suggested that the interdomain interaction change should be vital for signaling. However, the dynamics has not been clarified yet. In this study, we measured the photoreaction by the time-resolved transient grating (TG) method. We have detected several reaction phases which accompany absorption changes and/or diffusion coefficient (D) changes, which should be relevant for the signal transduction of Cph1.

1Pos021 アメリカヤマゴボウ抗ウイルスタンパク質におよぼす糖結合の影響

Sugar binding effects on pokeweed anti-viral protein

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Pokeweed anti-viral protein (PAP) which is classified into Type 1 RIPs makes complexes with glucose, NAG and other mono-saccharides with molecular ratio of 1:1 or 1:2. N-glycosidase activity of PAP was enhanced and simultaneously some specific conformational changes were induced near the active site by the interaction with sugar. Trp208 essential for the enzyme activity approached more closely to Tyr123 constituting the active center and furthermore, own rotational freedom was increased. The binding site of PAP for sugar was searched by the multi-dimensional FRET using two Trp-residues as the energy donor and synthesized fluorescent derivative of sugar as the acceptor. Based on the obtained results, the regulation mechanism of PAP by sugar is discussed in detail.

1Pos022 密度汎関数法による D-アミノ酸酸化酵素のフラビン-基質配置に関する研究

A DFT study on flavin-substrate arrangement in D-amino acid oxidase

Kyosuke Sato (*Dept. Mol. Physiol., Facult. Life Sci., Kumamoto Univ.*)

D-amino acid oxidase (DAO) is a flavoprotein that oxidizes D-amino acids (H_2N-CH(R)-COO^-) to form the corresponding imino acids (H_2N^+=C(R)-COO^-). Previously reported crystal structures of DAO complexed with substrate/product show that the C_α of D-amino acid is located near the N5 atom of flavin, indicating that the $_\alpha$H is transferred to the N5 atom as a hydride. In addition, the N atoms of substrate is located near the C4a atom of flavin. For the purpose of clarifying the significance of this flavin-substrate arrangement to the hydride transfer, the transition state energies were calculated with DFT for various arrangements. The result showed that the transition state energy is minimized when the flavin and substrate are arranged as in DAO.

1Pos023 26s プロテアソームの高速 AFM 観察

High-speed AFM observation of the 26S proteasome

Takashi Okuno[1], Kentaro Noi[2,5], Ken-ichi Arita-Morioka[2,5], Hikaru Tsuchiya[3], Yasushi Saeki[3], Kazunobu Takahashi[4], Tomonao Inobe[4], Takahiro Saito[1], Kunitoshi Yamanaka[2,5], Teru Ogura[2,5] ([1]*Fac. Sci. Univ. Yamagata.*, [2]*Inst. Mol. Embryol. Genet., Kumamoto Univ.*, [3]*Tokyo Metro. Inst. of Med. Sci.*, [4]*Frontier Res. Core for Life Sci., Univ. of Toyama*, [5]*CREST, JST*)

The 26S proteasome is a eukaryotic ATP-dependent protease responsible for selective and rapid degradation of polyubiquitinated substrate proteins. The 26S proteasome unfolds and translocates substrate into a proteolytic chamber through a narrow central pore. Direct observation of the substrate degradation reaction steps will provide new insights into the molecular mechanisms. We have analyzed the 26S proteasome from *Saccharomyces cerevisiae* by HS-AFM. The shape and size of the particle images obtained by HS-AFM resemble those resolved by cryo-EM. Dynamics of proteasome-substrate complexes using several designed model substrates is under intensive investigation with HS-AFM. We will discuss these results.

1Pos024 Structural origins of slowness and regulatory mechanism in KaiC ATPase

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The ATPase activity of KaiC is maintained always low but rhythmically fluctuated in the presence of KaiA and KaiB with the circadian period. To elucidate the regulatory mechanism of this unique ATPase, we have carried out structural and functional studies of KaiC and its mutants.
We will report two major findings, each of which was carefully evaluated by examining the crystal structures and the ATPase function. One finding is a potential structural-origin of slowness associated with ATPase. The other is a structural change effecting the fluctuation of the ATPase activity. These findings indicate that the circadian period is controlled and finely tuned through the structural changes accompanied with KaiC ATPase.

1Pos025 Study of interaction between transferrin and transferrin receptor2 by molecular simulation and flow cytometry

Tetsuya Sakajiri[1], Katsuya Ikuta[2], Takaki Yamamura[1] ([1]*Morioka Univ.*, [2]*Asahikawa Med. Col.*)

Mammals have 2 kinds of receptors (TfR1 and TfR2) of the iron transporter transferrin (Tf). Both TfRs share similar structures but possess different functions. TfR1 mediates cellular iron uptake by endocytosis. TfR2 is involved in the production of hepcidin which regulates iron homeostasis. The crystal structure of Tf-TfR1 has been reported. But, there is no report concerning the TfR2 structure. In this study, we created a 3D structural model of Tf-TfR2 and carried out a MD simulation to estimate bond energies of residual pairs between the Tf and the TfR2. The residual pair (Tf K144-TfR2 S138) was confirmed to be important using TfR2 mutants by a flow cytometry analysis (FCM). The FCM analysis supported importance of the residual pair predicted in the model structure.

1Pos026 植物型フェレドキシンの酸化還元電位を大幅に上昇させる構造要因の精密解析
Structural basis for the unexpected increase of redox potential of plant-type Ferredoxin revealed by the high resolution X-ray analysis

Daiki Kiyota[1,2], Arisa Sato[3], Risa Mutoh[2], Haruki Yamamoto[2], Toshiharu Hase[1,2], Genji Kurisu[1,2] ([1]*Grad. Sch. Sci. Osaka Univ.*, [2]*Institute for Protein Research, Osaka University, Japan*, [3]*Doshisha Women's college*)

Plant-type Ferredoxin (Fd) with a [2Fe-2S] cluster plays an important role in photosynthesis showing a very negative redox potential (-420 mV). Based on many structural and functional studies on Fd, a hydrogen bond network around the cluster is believed to be important to keep the redox potential negative. Unexpectedly, a single mutation of S45G (maize numbering) gave a drastic increase of redox potential (+180 mV). In order to understand how Fd tunes the redox potential precisely, we applied high-resolution X-ray structure analysis on Fd from Chlamydomonas reinhardtii. We solved the crystal structure of Fd at 0.89 Å resolution. Based on the high-resolution X-ray structure, we will discuss the reason why Fd shows the drastic increase upon mutation.

1Pos027 蛋白質多重配列アラインメントの格子気体模型
Lattice gas model of protein multiple sequence alignment

Akira Kinjo (*Inst. Protein Res., Osaka Univ.*)

The multiple-sequence alignment (MSA) of a protein family provides a wealth of information in terms of correlation patterns of residues at and between alignment sites. I have derived a lattice gas model of MSA based on the principle of maximum entropy with the constraints to satisfy observed statistics. The partition function, obtained by the transfer matrix method with a mean-field approximation, accounts for all possible alignments with all possible sequences. The model parameters for bonded and non-bonded interactions are determined by a self-consistency condition and by a Gaussian approximation, respectively. I apply this model to the globin and V-set domains by exerting global as well as local perturbations, and study the effect of long-range interactions.

1Pos028 Analysis of the Free Energy Landscapes for the Open-Closing Dynamics of MalK2 Using Enhanced Sampling MD Simulation

WeiLin Hsu, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech.*)

Protein dynamics are considered significant for several cellular processes. Due to their flexibility, proteins may stay in different states with or without the existence of the substrates. To describe the phenomena, two models have been proposed: the “induced fit” and the “conformational selection” mechanisms. Here, we used MalK2, the subunits of the E. coli maltose transporter, to understand its dimerization mechanism. Accelerated MD (aMD) and conventional MD (cMD) have been performed. The results revealed that the Mg-ATP binding led to a significant change in the free energy profiles, and thus stabilized the closed conformation. The free energy change caused by the ligand-binding indicates that the MalK2 dimerization would occur through the induced-fit mechanism.

1Pos029 How do intramolecular hydrogen bonds within a peptide contribute to peptide-antibody interaction?

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Protein-peptide interactions are common in living cells. But how do peptides overcome the loss of conformational entropy upon binding to their targets? To address this question, we analyzed the interaction between a peptide and an anti-peptide antibody. X-ray crystallography of the peptide-antibody complex revealed critical intramolecular hydrogen bonds (IHBs) within the peptide. Mutagenesis and isothermal titration calorimetry demonstrate that IHBs are formed upon binding, generating a favorable change of enthalpy. Interestingly, MD simulations indicate that IHBs in the peptide are formed transiently in solution before binding. Collectively, our results suggest that to minimize the loss of entropy the antibody recognizes a pre-configured peptide structure in solution.

1Pos030 Molecular dynamics of channelrhodopsin at the early stages of channel opening

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Channelrhodopsin (ChR) is a light-gated cation channel that responds to blue light. We performed electrophysiological analyses and all-atom MD simulations using recently solved crystal structure of a chimeric ChR, to investigate the importance of the intracellular and central constrictions of the ion conducting pore observed in the crystal structure of ChR. Moreover, we modeled the 13-cis retinal bound ChR and performed MD simulations to investigate the conformational changes in the early stage of the photocycle. Our simulations suggested that retinal photoisomerization induces the conformational change toward channel opening, including the movements of TM6, TM7 and TM2.

1Pos031 滴定 X 線小角散乱測定による Shootin1-Cortactin 複合体形成の構造学的評価

Structural investigation of direct interaction between Shootin1 and cortactin by the titration SAXS measurements

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Shootin1 is a clutch protein to mediate mechanical coupling between actin filaments and cell adhesions. It was reported that a F-actin binding protein, cortactin, interacts with Shootin1 by biological assays. In this study, we carried out SAXS measurements on shootin1-cortactin system to investigate the interaction between these proteins. We found that cortactin and shootin1 exist as a monomer and a dimer in solution, respectively. Cortactin-titration SAXS measurements upon shootin1 showed increase in inter-particle interference between shootin1 and cortactin. The binding stoichiometry is estimated to be 2:2 by the titration curve, indicating that cortactin is directly bound to shootin1. The biological assays are confirmed by solution structure assay.

1Pos032 Proton transfer mechanisms of photosystem II: Hybrid ab inito quantum mechanics study

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In photosystem II (PSII), two water molecules are progressively oxidized by electron transfer coupled to proton transfer driven by photons, where the protons and electrons are expelled from the catalytic site (i.e., the Mn cluster). In this study, we theoretically investigated the mechanisms of the proton transfer, which has still been open to be discussed. Employing hybrid ab inito quantum mechanics (QM)/molecular mechanics (MM) molecular dynamics (MD) simulations, we identified a novel pathway for the proton transfer where a Cl ion binding site in the vicinity of the Mn cluster is involved. Interestingly, this pathway includes a peptide group of Asn338 in the D1 domain, thus indicating that the amide can dislocate H+ depending on the environmental structure.

1Pos033 分子動力学シミュレーションを用いた細菌機械受容チャネル MscL の脂質膜厚変化で影響される開口過程の解析

Molecular Dynamics Study on the Gating of the Bacterial Mechanosensitive Channel MscL Affected by Membrane Thickness

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One of mechanosensitive channels MscL is homopentamer with two transmembrane inner and outer helices and gated by sensing membrane tension. Previous studies have shown MscL opens more easily when embedded in thinner bilayer, however, that remains unclear in detail. In this study, we performed MD simulations for the opening of MscL in the membrane with different thickness. As a result, MscL in thinner bilayer could open more widely. The interaction between lipids and MscL in thinner membrane became unstable in the initial step of the simulation and the lipid molecules around MscL were more orderly in thinner membrane even after membrane stretch, suggesting that MscL activates easily in thinner membrane due to more effective transmission of the force from the membrane.

1Pos034 Structure and dynamics of Sec protein-conducting channel

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Translocation of secretory proteins through the membrane is one of the evolutionally conserved mechanisms. We perform structural biological analyses for elucidation of protein translocation via SecYEG translocon, a conserved protein-conducting channel. Because the SecYEG complex is itself a passive channel, the driving force is required. SecA ATPase, a SecYEG-associated cytosolic motor, repeatedly pushes the preprotein into SecYEG channel using the energy of ATP hydrolysis. The conformational changes and interactions of Sec proteins during the protein translocation still remain unclear. Here we report new structures and model of Sec translocon. And we present the preparation of the reconstituted one unit of Sec machinery for visualization of the translocation process.

1Pos035 含セレン酵素の活性中心を模倣した短鎖セレノペプチドの分子設計

Molecular design of short selenopeptides mimicking a selenoenzyme active center

Natsuki Babe, Toshiki Suzuki, Taku Shimosato, Toshiya Minezaki, Michio Iwaoka (*School Sci., Tokai Univ.*)

Glutathione peroxidase (GPx) is an antioxidant enzyme that contains a selenocysteine residue at the active center. Although the existence of a catalytic tetrad, i.e., Sec(U), Q, W and N, is suggested in the GPx active site, the roles played by this tetrad have not been well established. Therefore, we have designed herein short selenopeptides with various amino acid sequences that can reasonably reproduce the GPx active site. Starting from a primitive sequence of GQAUAWNG, in which A is a spacer, we exchanged the essential amino acids Q, U, W, and N, with each other and mutated the spacer. The results of the SAAP-MC simulation, which is originally coded in our laboratory based on the SAAP force field, suggested RQPUPWNG can be an effective model of the catalytic tetrad.

1Pos036 内部座標系による構造エントロピー変化の計算と解析

Analysis for configurational entropy change calculated using internal coordinate system

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Configurational entropy (S_c) is an important component of free energies in biomolecular processes. We have assessed the accuracy of several calculation methods of S_c from molecular dynamics simulation by comparison with results of the Clausius method capable of accurately estimating the S_c changes with temperature (ΔS_c).

We found that the internal coordinate system is more suited to the quasi-harmonic approximation (QHA) for ΔSc than the Cartesian coordinate system. Analyses of the components of ΔS_c indicate that 1D entropy terms dominantly contribute to ΔS_c, and that anharmonic-entropy changes arise from the changes of probability distributions of torsional angles.

1Pos037 超音波連続照射下におけるマウスプリオン蛋白質の凝集体形成

The formation of aggregates of mouse prion protein under the continuous ultrasonic irradiation

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Although the ultrasonic irradiation induces the formation of amyloid fibrils *in vitro*, the details of aggregation occurring under the ultrasonic irradiation are unknown. To examine the conformational change of mouse prion protein (mPrP), we measured the far-UV CD along with the continuous ultrasonic irradiation. The result indicated that the CD ellipticities significantly decreased within 10 minutes at around its isotropic point (pI) of 9.6. EM observation showed that small oligomers and large amorphous-like oligomers were formed at acidic pHs and around its pI, respectively. Thus, the continuous ultrasonic irradiation will disrupt the supersaturation state and be a trigger for the formation of oligomers or aggregates of mPrP.

1Pos038 SyPixD の光反応に対する構造揺らぎの効果

Effect of structural fluctuation on photoreaction of SyPixD

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We investigated the relevance of structural fluctuation to the protein's reaction of SyPixD, which is a blue light sensor containing a BLUF domain. SyPixD forms the decamer in solution in the dark state and dissociates into the dimers upon excitation of the two monomer units in the decamer. This dissociation does not take place upon one monomer unit excitation. For monitoring the relationship between the reaction yield and the structural fluctuation of reaction intermediates, we measured the compressibility change during the photoreaction of SyPixD by transient grating method. We found that the fluctuation of the intermediate decreased when the light intensity for the excitation was decreased, which may explain the light intensity-dependent reaction yield.

1Pos039 アルコール脱水素酵素のサブユニット間相互作用に及ぼすホフマイスター効果

Hofmeister effect on the subunit-subunit interaction of Liver Alcohol Dehydrogenase

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Hofmeister effect is a clue to elucidate the ion effects on various biological phenomena. We reported last year that the domain-domain interaction of GFP-GST fusion protein was regulated by monovalent cations in following to Hofmeister series. In the present study, the subunit-subunit interaction was examined using energy homo-transfer between Trp residues in the dimeric form of LADH. The fluorescence depolarization ratio ($r_{ex315nm}$/$r_{ex295nm}$) which is a criterion for the degree of energy homo-transfer increased according to the ion species and concentration to suggest that the regulation of subunit-subunit interaction would be due to Hofmeister effect. These results were further confirmed by the time-resolved fluorescence studies of the energy homo-transfer between Trps.

1Pos040 ユビキチン化の新規物理化学的性質

若手招待講演

Novel physicochemical properties of ubiquitylation

Daichi Morimoto[1], Erik Walinda[1], Kenji Sugase[1], Harumi Fukada[2], Yu-shin Sou[3], Shun Kageyama[3,4], Masaru Hoshino[5], Takashi Fujii[6], Hikaru Tsuchiya[7], Yasushi Saeki[7], Kyohei Arita[8], Mariko Ariyoshi[1], Hidehito Tochio[9], Kazuhiro Iwai[10], Keiichi Namba[6,11], Masaaki Komatsu[3,4], Keiji Tanaka[7], Masahiro Shirakawa[1] ([1]*Eng., Kyoto Univ.*, [2]*Life Envi. Sci., Osaka Pref. Univ.*, [3]*Protein Metabolism Proj., Tokyo Metro. Ins. Med. Sci.*, [4]*Med., Niigata Univ.*, [5]*Pharm., Kyoto Univ.*, [6]*QBiC, RIKEN*, [7]*Lab. Protein Metabolism, Tokyo Metro. Ins. Med. Sci.*, [8]*Med. Life Sci., Yokohama City Univ.*, [9]*Science, Kyoto Univ.*, [10]*Med., Kyoto Univ.*, [11]*Frontier Biosci., Osaka Univ.*)

Ubiquitin is one of the most stable proteins, but it is often found in inclusion bodies associated with neurodegenerative diseases such as Alzheimer's disease. To gain insight into this contradictory behavior, we examined the physicochemical properties of ubiquitin and its polymeric chains. We found that the folding stability of ubiquitin chains decreased with increasing chain length, resulting in amyloid-like fibril formation. When expressed in cells, polyubiquitin chains also formed aggregates depending on chain length. Notably, these aggregates were selectively degraded by autophagy. We propose that the instability of polyubiquitin chains drives fibril formation, which serve as an initiation signal for autophagy.

1Pos041 時間分解 EPR 法を用いたアミロイド線維形成過程におけるヒトインスリン局所構造変化の観測

Time-resolved EPR Study on Local Structures of Human Insulin Forming Amyloid Fibrills

Tomoka Abe, Takashi Tachikawa, Eri Chatani, Yasuhiro Kobori (*Grad. Sch. Sci., Kobe Univ.*)

Protein aggregates produced by the misfolding are called amyloid fibrils. It has been known that the amyloid fibrils are causes of many serious diseases. Since any resolutions of these diseases have not been discovered, it is of great importance to clarify how these amyloid fibrillations occur. We have employed human insulin as a model protein and anthraquinone derivative to generate a photoinduced charge-separated state in the protein complex. We have characterized the protein-ligand structured for the native states and for the fibril states by the time-resolved EPR method. Different spectra have been obtained by the EPR experiences between native states and fibril states. We have predicted the possible structural change of the protein's local structure from spectrum.

1Pos042 膜貫通 β バレルの β ストランドのねじれと曲りは抑制されている

β-strand twisting and bending of the transmembrane β-barrel are suppressed

Nobuaki Kikuchi, Shinichi Ebisawa, Yuka Watanabe, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. of Bioinformatics, Soka Univ.*)

It is well known that the majority of β-sheets consists of the twisted and the bended β-strands. In the previous study for water soluble proteins, Ser, Thr and Asn side-chains were found to suppress twisting β-strand. In this study, we focused on the twisting and bending in β-barrel transmembrane proteins and analyzed frequency distributions of twist and bend angles of β-strands in known structures. The results showed that the most frequent twist and bend angles of β-strands in membrane proteins were smaller than those in water soluble proteins, respectively. This tendency was also observed even though β-strands have no Ser, Thr or Asn residues, suggesting the existence of another twist-suppressing factor in membrane proteins.

1Pos043 複数のアミロイド性ペプチドの混在する複雑な系におけるアミロイド線維形成機構
Amyloid Fibrillation in Complicated Systems Containing Various Amyloidogenic Peptides

Hiroya Muta[1], Masatomo So[1], Kazumasa Sakurai[2], Yuji Goto[1] ([1]*Inst. for Pro. Research, Osaka Univ.*, [2]*Inst. of Adv. Tech., Kinki Univ.*)

Amyloid fibrils form in supersaturated solutions via a nucleation and growth mechanism. Although there are many studies addressing amyloid fibrillation of single amyloidogenic protein or peptide, amyloid fibrillation under complicated systems containing various proteins has been far from clear. As a model of amyloid fibrillation under a complicated system, we compared the fibril formation of K3 fragment of β2-microglobulin produced by Achromobacter protease I with that in a mixture after proteolysis. Solution NMR combined with amyloid specific thioflavin T fluorescence revealed that the interactions of K3 with other peptide fragments retard fibrillation, suggesting the mechanism of amyloid fibrillation in complicated systems, especially under physiological environments.

1Pos044 Study on the NaCl concentration-dependent structural stability of maize (leaf) ferredoxin

Misaki Kinoshita, Yoko Ariga, Toshiharu Hase, Yuji Goto, Genji Kurisu, Young-Ho Lee (*Institute for Protein Research, Osaka University*)

Acidic proteins of halophilic bacteria have been shown to increase their conformational stability with increasing the concentration of salts; however, this halophilic property of ferredoxin (Fd) remains unknown. We examined the changes in thermal stability of maize leaf Fd at various NaCl concentrations. Circular dichroism spectroscopy and differential scanning calorimetry indicated that the increases in NaCl concentrations enhanced the thermal stability of Fd by showing increases in Tm and enthalpy changes. The similar findings were also detected using the hydrogen-deuterium exchange method and NMR spectroscopy. We suggested that the decrease in the flexibility of Fd and the increase in hydrophobicity in cores at high NaCl concentrations increased the stability of Fd.

1Pos045 抗がん剤候補のハースタチンは天然変性タンパク質である
Herstatin, an antitumor drug candidate, is an intrinsically disordered protein

Daisuke Tashiro, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

A receptor tyrosine kinase HER2 forms a dimer to promote cell proliferation, but overexpression of HER2 causes cancers. Therefore, a HER2-binding protein that blocks its dimerization is expected as an antitumor drug. A HER2 alternative splicing product, herstatin, is an autoinhibitor of HER2, and the C-terminal, intron 8-encoded domain of herstatin (Int8) is known to bind HER family receptors. Here, we show by CD, NMR, and X-ray scattering that Int8 is intrinsically disordered. CD and HSQC spectra reveal that Int8 is largely disordered but with residual helical structures. A radius of gyration of Int8 is in between those of the typical molten globules and fully unfolded states. These results demonstrate that herstatin is an intrinsically disordered protein.

1Pos046 GB1(41-56)ペプチドのβヘアピン構造に及ぼす圧力効果に関するFTIR・ラマン分光研究
FTIR and Raman studies of pressure effects on the β-hairpin structure of GB1(41-56) peptides

Minoru Kato[1], Keita Tsuchiya[2] ([1]*Dept. Pharmacy. Ritsumeikan Univ.*, [2]*Grad. Sch. Life Sci. Ritsumeikan Univ.*)

For understanding the mechanism of pressure unfolding of proteins, recently we have been performing systematic studies using model peptides. In the preset study, we focus on the β-hairpin structure, which is the simplest β-structure. The current targeted peptides of research are mutants for GB1segment peptides, which are composed of residues 41-56 of the B1 domain of protein G (GB1). These β-hairpin structures are well known to be stabilized by the hydrophobic interactions between W43 and V54, and between Y45 and F52. We have reported pressure effect on the β-hairpin structure of triple mutants of GB1 segment peptide (W43Y45F52V54) via FTIR and Raman spectroscopies, which provide the structural information of the peptide backbone and hydrophobic cluster, respectively.

1Pos047 Control of protein aggregation and oligomerization using short SEP (Solubility Enhancing Peptide) tags

Md. Golam Kabir, Mohammad Monirul Islam, Yutaka Kuroda (*Tokyo Univ. Agri. Eng.*)

We report the effects of short peptide tags, consisting of 5 consecutive single type amino acids attached to the C terminal end of a protein, on the formation of sub-visible soluble oligomers, using a bovine pancreatic trypsin inhibitor variant as a model. Dynamic light scattering, at pHs 4.7-8.7, indicated that SEP tags composed of K, D and E favored 3-4mers; whereas S, N, T and Q tags favored dimers. The hydrophobic I and L containing tags favored the formation of 7-15mers, together with large insoluble aggregates. These results were fully corroborated by size-exclusion chromatography experiments, except for the C5I and C5L tagged variants. These observations clearly suggest that SEP tags can be used for controlling the formation of sub-visible soluble oligomers.

1Pos048 ダイナミン 1-コルタクチンおよびダイナミン 1–アンフィファイジン複合体の高速 AFM による動態観察
High-Speed AFM imaging of dynamics of Dynamin 1-Cortactin and Dynamin 1-Amphiphysin 1 complexes

Yusuke Kumagai[1], Tetsuya Takeda[3], Takayuki Uchihashi[1,2], Tadashi Abe[3], Kohji Takei[3], Toshio Ando[1,2] ([1]*College of Science and Engineering, Kanazawa University*, [2]*Bio-AFM Frontier Research Center, College of Science and Engineering, Kanazawa University*, [3]*Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama University*)

Dynamin 1 is a GTPase involved in diverse processes including actin dynamics and endocytosis. Dynamin 1 forms ring-shaped complexes with either Cortactin or Amphiphysin 1 and these complexes are thought to function in actin bundling and membrane scission, respectively. Several electron microscopic studies showed that these Dynamin 1 complexes dynamically change their structures upon GTP hydrolysis. However, dynamics occurring in these Dynamin 1 complexes has remained elusive. Here, we applied high-speed AFM to observe GTP hydrolysis-coupled dynamic changes of these protein complexes directly. In the presentation, we will show recent results of these observations.

1Pos049 線虫ミトコンドリアにおけるフェレドキシン依存性電子伝達経路の機能解析

Characterization of ferredoxin-dependent electron transfer system in Caenorhabditis elegans mitochondira

Ryohei Wada[1], Hiroshi Hori[1], Yoshito Furuie[1], Fusako Takeuchi[2], Motonari Tsubaki[1] ([1]*Grad. Sch. Sci., Chem., Kobe Univ.*, [2]*IPHE., Kobe Univ.*)

In mitochondria, two types of ferredoxin (Fd) exist. Fd-1 involves in steroidogenesis where electrons are transferred from NADPH via Fd reductase (FdR) to Fd-1, which in turn reduces CYP. Fd-2 contributes to Fe-S cluster assembly. However *C. elegans* possess Fd-2 only, despite of the presence of mitochondrial CYP. To elucidate the electron transfer system, we tried to reconstruct the system using CeFd and CeFdR proteins expressed by *E. coli*. The UV-vis and EPR spectra of recombinant CeFd protein indicated the presence of [2Fe-2S] cluster with an Fd-2-type character. For the expression of CeFdR, 3% ethanol was added to the medium after induction with IPTG. Cyt*c* reduction assay indicated that CeFdR has an electron transfer activity through Fd-1.

1Pos050 ラン藻でのアルカン合成に必要な 2 つの酵素間の結合部位の探索

The search for the binding sites between two enzymes essential for cyanobacterial alkane biosynthesis

Mari Chang[1], Yuuki Hayashi[2], Munehito Arai[1,2] ([1]*Dept. Phys., Univ. Tokyo*, [2]*Dept. Life Sci., Univ. Tokyo*)

Cyanobacterial biosynthesis of alkanes is catalyzed by a two-step reaction involving acyl-(acyl carrier protein) reductase (AAR) and aldehyde deformylating oxygenase (AD). Recently, both enzymes are reported to interact with each other for efficient delivery of aldehyde from AAR to AD. To search for the AAR-binding site on AD, here we carried out an alanine scanning mutagenesis around the substrate-entry/exit site of AD, and have found that Ala substitutions at 8 residues greatly reduced the alkane production in *E. coli* coexpressing AAR and AD. Four out of the 8 residues are located at the surface of the crystal structure of AD, suggesting the AAR-binding site. *In vitro* binding assay by gel filtration of the mixture of AAR and the AD mutants is currently underway.

1Pos051 アルカン合成酵素のアラニンスキャン変異解析

Residues essential for the alkane producing activity of aldehyde deformylating oxygenase revealed by alanine scanning mutagenesis

Keigo Shimba, Fumitaka Yasugi, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

Alkane biosynthesis by cyanobacteria is an attractive way of producing renewable substitutes for fossil fuels. The final step in the pathway is the conversion of aldehydes into alkanes by aldehyde deformylating oxygenase (AD). Crystal structures of AD have suggested a reaction center and a substrate-binding channel. However, the issues of which residues in the active site are catalytically more important and whether other residues far from the active site are also essential for the catalysis remain unknown. To clarify these issues, we performed alanine scanning mutagenesis of AD, especially at conserved residues. We found that the residues comprising the iron binding sites are indispensable for the catalysis. Roles of the conserved residues in AD will be discussed.

1Pos052 海洋性ビブリオ菌のべん毛本数を負に制御する FlhG の蛋白質凝集性と ATPase 活性の相関

Relationship between aggregability and ATPase activity of FlhG, the negative regulator of the flagellar number in *Vibrio alginolyticus*

Hikaru Hirata, Michio Homma, Seiji Kojima (*Div. Bio Sci., Grad. Sch. Sci., Univ. Nagoya*)

Marine bacterium *Vibrio alginolyticus* has a single polar flagellum whose number is regulated negatively by FlhG. FlhG is a homolog of MinD, a cell division inhibitor of Escherichia coli. FlhG has a low ATPase activity and the ATPase motif is necessary for proper function of FlhG. Here we investigated the size of FlhG molecule by dynamic light scattering and by gel filtration chromatography. We show that FlhG exists as a monomer even upon hydrolyzing ATP, which is distinct from MinD. Moreover, we found that a mutant conferring high ATPase activity aggregated in the presence of ATP. This mutant also exhibited high regulatory function of FlhG. These results suggest that FlhG changes its conformation upon ATP hydrolysis, which is important for its function.

1Pos053 4. Development of a new BRET based ATP indicator for high-throughput quantitative ATP assay

Tomoki Yoshida, Hiromi Imamura (*Graduate school of biostudy, Kyoto University*)

ATP is an important component as the energy currency in cells. In this study, we developed a new ratiometric ATP indicator using bioluminescence resonance energy transfer (BRET) principle. The indicator, named BTeam, was composed of the ε subunit of the bacterial ATP synthase, ATP-independent lusiferase and yellow-fluorescent proteins (YFP). BTeam showed BRET changes along with ATP levels, which were evaluated by YFP emission per lusiferase emission. A decrement in the ATP levels inside HeLa cells was observed after treatment with inhibitors of glycolysis and oxidative phosphorylation by using BTeam. BTeam would allow a high-throughput quantitative ATP assay by monitoring the levels inside living cells with only addition of lusiferin to the medium.

1Pos054 難溶性化合物とタンパク質との相互作用解析へ向けた新規技術開発

A novel approach for analysis of interaction between low water-soluble compounds and target proteins

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The structural information of the protein-ligand complex has the potential to find ways to improve the lead compounds. The best way is to determine the 3D structure of the complex by soaking the ligand in crystals. However, the soaking step has a problem because many lead compounds are low water-soluble. Such lead compounds must be dissolved in concentrated organic solvents, such as DMSO. We recently developed a new method for growing crystals in a high-strength hydrogel. This method enabled us to increase the mechanical stability of the crystals. In this study, we transferred the hydrogel-grown avidin or FABP crystals into a 50% DMSO solution containing the inhibitors. We observed the clear electron density maps of the compounds that are bound to the active sites.

1Pos055 X 線 1 分子追跡法よるフェムトニュートン力場でのタンパク質ネットワーク観察

Observation of Supersaturated Protein Networking in femto-Newton Force-field from Diffracted X-ray Tracking

Yufuku Matsushita[1], Hiroshi Sekiguchi[2], Noboru Ohta[2], Keigo Ikezaki[1], Yuji Goto[3], Yuji C. Sasaki[1,2] ([1]*Graduate School of Frontier Science, The University of Tokyo*, [2]*SPring-8/JASRI*, [3]*Institute for Protein Research, Osaka University*)

The supersaturation is widely known to be a precursor to the crystallization, precipitation and aggregation process in inorganic, organic, polymer and protein solution systems. In this study, we tried to observe local dynamical structures of metastable crystallization of lysozyme solution by Diffracted X-ray Tracking as a high positional accuracy (-pm) and time resolved (100 us) detection system. From this experiment result, we confirmed that this condition is consisted by the different dynamics modes (Fast and Low). Then, from detailed analysis of fast dynamics mode, we observed the localized extremely small force (fN scale) existence as a relaxation process of characteristic phenomena in supersaturated condition.

1Pos056 アカネ科植物由来 抗腫瘍活性ペプチド RA-VII の分子動力学シミュレーション

Molecular dynamics simulations of antitumor peptide RA-VII from Rubia cordifolia

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RA-VII is a cyclic hexapeptide, which is isolated from Rubia cordifolia and Rubia akane. This peptide shows potent antitumor activity, and comprised of alanines and modified tyrosines. The peptide has an 18-membered cyclohexapeptide ring and a 14-membered cycloisodityrosine ring in the structure. It was reported that the peptide has two or three conformations in solvents. In the NMR experiments, the peptide in DMSO-d6 showed different conformational behavior from that in other solvents; the signals of the third conformer, not detected in chloroform-d or in dioxane-d8, was appreciably observed in this solvent. We studied the conformers of RA-VII and its analogs by molecular dynamics simulations to analyze the structure of RA-VII in DMSO.

1Pos057 解析ソフトウェア CAPAXIS のさらなる開発：PyMOL との連携と B 型肝炎ウイルスへの応用

Further Development of Software Tool, CAPAXIS, for Use with PyMOL : Application to Hepatitis B Virus

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CAPAXIS is useful software to analyze virus capsid structures on the basis of icosahedral symmetry. CAPAXIS has functions such as classification of rotation axes, calculation of cell numbers, generation of the entire structures and superposition of capsid structures to different coordinate axes. In this study, PyMOL scripts and plugins were developed to use CAPAXIS associated with the molecular visualization system, PyMOL. New functions were also added into CAPAXIS for generating arbitrary symmetry-related cell structures and adding water molecular around capsid in the cell for simulation in the rotational symmetry boundary condition. We applied the developed CAPAXIS to molecular dynamics simulation of hepatitis B virus.

1Pos058 新型マイクロアレイ MMV を用い機能性ペプチドを淘汰するのに初めて成功したーセルベーストスクリーニングの躍進

First success in selection of functional peptides using a novel concept microarray MMV－A breakthrough for cell-based screenings

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Ultra-HTS is most wanted by various fields of scientists due to the bursting number of data (leading to higher cost) required in omics studies, combinatorial sciences, and so on. Usually, it has been pursued by robotics. Here, a quite different approach was developed. As model cases, functional peptides for regulating proteins, p38α,CREB, and MGMT were hunted by the MMV-based screening technology (Sharma et al. BMC Biotech. 2014). Peptides of an inhibitory function against MGMT were newly selected within two days. This success can be directly connected to the applicability of this method to cell-based screening for various purposes. In principle, most of experiments performed by using microplates were shown to be replaced with MMV-based methods.

1Pos059 主鎖二面角パターンによる αβ 型タンパク質構造のデザイン

Design of αβ protein structures: the extended design principles

Yu-Ru Lin[1], Rie Koga[1,3,4], Gaohua Liu[2], Guy Montelione[2], David Baker[1], **Nobuyasu Koga**[1,3,4] ([1]*Univ. Wash.*, [2]*Rutgeres, NESG*, [3]*IMS, CIMoS*, [4]*JST, PRESTO*)

We recently described principles for designing ideal protein structures that have perfect consistency. The principles were based on a set of rules relating secondary structure patterns to tertiary motifs, in which loop and secondary structure lengths play critical roles in specifying desired tertiary structures. Here, to control over protein shape and size within a topology, we further developed the design principles by extending the rules to the level of backbone torsion patterns. We found that the loop geometries are classified by backbone torsion patterns. The four designed protein structures for two folds show close agreement with the NMR structures. The extended design principles provide a foundation for custom design of protein structures.

1Pos060 合理的設計による抗体精製用リガンド FPA の抗体解離 pH の向上

Rational design to improve the pH-sensitive antibody dissociation of FPA, a ligand for antibody purification

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Protein A (PA) is frequently used as an affinity ligand for antibody purification. However, acidic pH is required to dissociate antibodies from PA, which may cause immunogenic aggregation of antibodies. Therefore, to manufacture safe and secure antibody medicines, it is necessary to develop an affinity ligand that can dissociate antibodies at moderate pH. We have previously developed a novel affinity ligand, FPA, which is a fusion of PA and the coiled-coil region of c-Fos. A rationale for the design is given by “mutually exclusive binding”, in which the homodimeric coiled-coil formation of c-Fos at low pH dissociates an antibody from PA. Here, we rationally introduced multiple amino acid substitutions into FPA, and succeeded in dissociating antibodies at pH 5.1.

1Pos061 Rossmann フォールド蛋白質のデザイン：ストランド入れ替わり問題の謎に迫る

Design of Rossmann folds: the internal strand swapping problem

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We recently described general principles for designing ideal protein structures. Here, we applied the principles to the design of Rossmann2x3 fold consisting of five strands. However, the design folded into the same architecture but the different topology, Ploop2x3 fold, in which internal two strands are swapped. Why did the design fold into the different topology? We investigated naturally occurring Rossmann2x3 structures and found that the landing position of the 1-strand crossover helix in most of the naturally occurring proteins is shifted two residues from that of the design model. Based on the finding, we built the backbone structures again and designed amino-acid sequences, and finally we obtained a sequence that can fold into the Rossmann2x3 fold.

1Pos062 電気化学マイクロデバイスによる P450 反応解析

Electrochemical Analysis of P450s in Microfluidic Channel

Yasuhiro Mie, Yasuo Komatsu (*Bioproduction Res. Inst., AIST*)

Cytochrome P450 enzymes catalyse a vast array of oxidative biotransformations that are potentially useful for industrial synthesis and pharmaceutical analysis. Understanding and development of cost-effective assay for the P450 catalysis are important. Since P450 requires reducing power, electrochemical approach to analyse and apply the reaction for the purposes can be useful. Especially, the reaction in a microfluidic channel is expected to construct rapid screening system of the various P450 reactions. In the present study, we have examined the electrochemical reactions of microsomal and bacterial P450s in the microfluidic system with our nanostructured electrode system, and observed the catalytic signals corresponding to the reaction rate.

1Pos063 ヘムの構造歪みが酸化還元電位に与える影響の計算科学的研究

Computational Study of Structural Effects on Redox Potential of Hemes

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Hemoproteins, metalloproteins comprising heme groups, serve electron transfer, oxygen transport, and redox catalysis. The multifunctional characters of hemoproteins can be attributed to flexibility of the electronic and molecular structures of heme regulated by proteins. The relationship between the property and structure of a simple heme leads to the mechanism for the multifunction of hemoproteins.

In order to understand the structure-property relationship of hemes, we constructed distorted heme models by Normal-coordinate Structural Decomposition (NSD) method, which expresses a heme distortion as a linear combination of vibrational modes, and we evaluated the redox potentials with QM calculations. The importance of an in-plane distortion has been found.

1Pos064 Bach2 天然変性領域の電荷状態分布の解析

Charge-state-distribution analysis of Bach2 intrinsically disordered heme binding region

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Bach2 is a transcriptional regulator, including two structural domains (BTB and bZip) and the intrinsically disordered region as the heme-binding sites which are known as the CP motif. It is shown that the charge-state-distribution (CSD) in the Electron-Spray-Ionization (ESI) mass spectroscopy is highly correlated with a protein structure in solution. To investigate conformational change by heme binding on Bach2, the wild type Bach2(331-520) and its CP-mutant (cysteine to alanine) were analyzed by ESI mass spectrometry. In this study, we analyzed the CSD of Bach2 with/without heme. In heme binding conditions, the most intense peak of CSD was shifted to the lower charge state. This result suggests that heme induces more compact form of Bach2.

1Pos065 チトクロム酸化酵素を用いた常温高分解能 X 線回折実験法の確立

Development of high-resolution X-ray diffraction experiments at room temperature

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Cytochrome *c* oxidase (C*c*O) reduces O_2 to H_2O coupled with proton pumping. Freezed crystals are used for X-ray diffraction experiment to reduce radiation damage. However a structure of C*c*O at cryogenic temperature is not always the same as that in vivo. Thus we have to establish an experimental procedure at room temperature. HAG method developed by Baba *et al.* was applied to the experiment of C*c*O at room temperature. A crystal was coated with polyvinyl alcohol solution and exposed to X-rays at BL38B1 of SPring-8 under the humidity-controlled condition at 293 K. It diffracted up to 3.0 Å, and its lattice constants and mosaicity were not changed during 1 h X-ray exposure. We are currently applying the method at 278 K to improve its resolution.

1Pos066 Electrochemical behavior of bacterial nitric oxide reductase immobilized on gold electrodes *via* self-assembled monolayers

Kuniaki Yamaki[1], Masaru Kato[1], Takehiko Tosya[2], Ichizo Yagi[1] ([1]*Grad. Sch. Env. Sci., Univ. Hokkaido*, [2]*Harima Inst., Riken*)

Nitric oxide reductase (NOR) is an enzyme, which catalyzes reduction of nitric oxide (NO) to nitrous oxide (N_2O) at a catalytic site containing iron atoms. We will report immobilization of bacterial NOR, which was isolated from *Pseudomonas aeruginosa*, on Au electrodes *via* NH_2- or COOH-terminated self-assembled monolayers and electrochemical catalytic activities of the immobilized NOR under catalytic/non-catalytic conditions. The orientations of the immobilized NOR controlled by the surface charges might give some effects on its electrochemical catalytic activities. We will also report spectroelectrochemical studies on the immobilized NOR.

1Pos067 固体 NMR によるフォボロドプシン-トランスデューサー複合体の細胞質側での相互作用の解析

Analysis of interactions of phoborhodopsin-transducer complex in cytoplasmic side by solid-state NMR spectroscopy

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Phoborhodopsin from *Natronomonus pharaonis*, *p*pR is a photoreceptor membrane protein with a retinal chromophore and forms 2:2 complex with the cognate transducer protein, *p*HtrII. It is important for understanding the signal transduction mechanism to investigate an interaction between *p*pR and *p*HtrII in the cytoplasmic region. In this study, we investigated the interaction by solid-state NMR experiments. We observed the difference of T_{NH} and ^{1}H $T_{1\rho}$ relaxation times of Arg residues located at cytoplasmic side between *p*pR alone and *p*pR- *p*HtrII complex. It is indicated that there is the interaction of Arg residues in *p*pR with *p*HtrII. We will adjust reconstituted condition of *p*pR to obtain high-resolution multi-dimensional solid state NMR spectra.

1Pos068 GPCR の熱安定性を向上させるアミノ酸置換の予測:有力な手法の構築

Prediction of Thermostabilizing Mutations for G Protein-Coupled Receptors: Construction of an Efficient Method

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Here we construct an efficient method of predicting thermostabilizing mutations for G protein-coupled receptors. We consider the adenosine A2A receptor under the postulation that its wild-type structure is unknown. Our method comprises the following steps: Construct candidate structures of the wild-type via homology modeling; select the best structure using our free-energy function (FEF); estimate the structures of a number of mutants by the MODELLER and choose the mutations leading to enhanced stability using our FEF; and experimentally examine if some of these mutations actually lead to enhanced stability or not. We find that the success rate is almost the same as that in the case where the wild-type structure is known and utilized.

1Pos069 腸内連鎖球菌 V-ATPase のクライオ位相差電子顕微鏡単粒子構造解析

Single particle phase-contrast cryo-EM 3D reconstruction of Enterococcus hirae V-ATPase

Kazuyoshi Murata[1], Takeshi Murata[2], Hiroshi Ueno[3], Ryota Iino[4] ([1]*National Institute for Physiological Sciences*, [2]*Dept. Science, Chiba University*, [3]*School of Engineering, The University of Tokyo*, [4]*Okazaki Integrated Bioscience/ Institute of Molecular Science*)

Eh V-ATPase is an ATP-driven Na+-pump isolated from Enterococcus hirae, which is consisted of 24 subunits from 9 different proteins. Here, we applied the Zernike-phase contrast cryo-electron microscope (ZPC-cryoEM) to take images of the whole complex of the detergent-solubilized Eh V-ATPase. A single particle 3D reconstruction of the ZPC-cryoEM images successfully revealed a whole structure of Eh V-ATPase at near 1 nm resolution. A fitting of X-ray crystallographic models of individual subunits showed a different state of ATP hydrolysis and a possible interaction between the ATPase motor (V1) and the Na+-pump (Vo). The density of inner membrane region suggested the molecular mechanism of Na+-pump.

1Pos070 High-resolution cryoEM structural analysis of MotPS stator of the bacterial flagellar motor

Naoya Terahara, Takayuki Kato, Tohru Minamino, Keiichi Namba (*Grad. Sch. of Frontier Biosciences, Osaka Univ.*)

The bacterial flagellar motor is powered by electrochemical potential difference of cations across the cytoplasmic membrane. Energy is provided by cation influx through the channel of the stator complex. Mechanism of rotation is unknown due to the lack of structural information of the stator. Here we report purification and cryoEM single particle image analysis of a sodium-type stator complex, MotPS. Purified MotPS complexes were mixed with amphipol and biobeads to remove the detergent for cryoEM grid preparation. The MotPS complex was stable and monodispersed in the detergent-free solution. Classification and average of cryoEM images revealed a tetrameric structure of the MotPS complex. For high-resolution 3D image reconstruction, further image analysis is in progress.

1Pos071 Highly sensitive measurement of proton flux mediated by POT

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Proton-dependent oligopeptide transporter (POT) is a MFS protein which drives the uptake of nutrient peptides by using the proton gradient across cell membrane (ΔpH). Although the transport activity of POT has been well studied using radioisotope-labeled peptides as substrates, it still remains difficult to analyze the peptide-coupled proton flux down the proton gradient due to low sensitivity. In this study, we attempted to measure the proton flux mediated by POT using an arrayed lipid bilayer chamber system (ALBiC), which allows to measure the proton flux with great sensitivity. When reconstituted with POT, ALBiC exhibits the POT-mediated proton flux as a fluorescent decrease of pH indicator, suggesting that the proton flux per POT is faster than 300 s^{-1} at ΔpH of 1.

1Pos072 メカノセンシティブチャネルはストレプトマイシンの細胞内への侵入に関与するか？

Are mechanosensitive channels involved in the entry of streptomycin into the bacterial cell?

Kenichi Hashimoto, Fangzhen Zheng, Kazuhiro Nobata, Sanae Yamazaki, Isamu Yabe, Hisashi Kawasaki (*Dept. Green Sust. Chem., Tokyo Denki Univ.*)

The primary mechanism of action for streptomycin is the inhibition of translation. Nonetheless, it is yet unknown how streptomycin enters the bacterial cell. In 2014, it was reported that the potency of streptomycin is dependent on MscL expression. MscL belong to a family of mechanosensitive channels (Msc), and the study suggests that streptomycin enters the investigated cell by activation of these Msc. Here, we investigated whether the Msc are influenced by streptomycin. In *Corynebacterium glutamicum*, it was revealed that the sensitivity to streptomycin is decreased in a strain with disrupted Msc. In addition, we obtained patch-clamp data that seems to suggest that streptomycin is capable of influencing Msc.

1Pos073 1分子イメージングで見るGPCRの多量体化とエンドサイトーシス

Single-molecule imaging of GPCR oligomerization followed by internalization

Masataka Yanagawa[1], Michio Hiroshima[2], Takahiro Yamashita[3], Yoshinori Shichida[3], Yasushi Sako[1] ([1]*Riken*, [2]*QBiC, Riken*, [3]*Grad. Sch. Sci., Kyoto Univ.*)

GPCR dimers have attracted much attention as potential drug targets because the dimerization regulates receptor functions including a ligand binding affinity, and G protein activation efficiency. However, a physiological importance of higher-order oligomerization of GPCRs is unclear, which cannot be distinguished from dimerization by conventional biochemical assays. Here we directly observed single-molecule dynamics of metabotropic glutamate receptor (mGluR), a prototypical class C GPCR, on the living cell surface by using total internal reflection fluorescence microscopy. The higher-order oligomerization of mGluR occurs with deceleration of the diffusion followed by the clathrin-dependent internalization, which is tightly coupled with receptor activation.

1Pos074 上皮成長因子受容体クラスタリングの超解像定量解析

A Super-resolved Quantitative Analysis of Epidermal Growth Factor Receptor Clustering

Michio Hiroshima[1,2], Masahiro Ueda[1], Yasushi Sako[2] ([1]*RIKEN QBiC*, [2]*RIKEN*)

Clustering of epidermal growth factor receptor (EGFR) has been suggested to be inextricably concerned with the activation of EGFR which evokes various cellular signaling. However, details in the clustering mechanism and the cluster conformation remain unknown. For a precise analysis of the clustering, here we employ photo-activation localization microscopy (PALM). The distribution of oligomer size was quantified by a hierarchical clustering algorithm on the super-resolved data and distinct from that obtained by a conventional microscopy and analysis. The improved localization accuracy enabled an investigation of the spatial receptor arrangement in oligomers. A mechanism of clustering regulation unveiled by the method will be discussed.

1Pos075 粗視化MD-SAXS法とヌクレオソームへの適用

Coarse-Grained MD-SAXS method and application to nucleosomes

Yuichi Kokabu[1], Takashi Oda[1], Masaaki Sugiyama[2], Hitoshi Kurumizaka[3], Mamoru Sato[1], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*, [2]*Grad. Sch. Sci., Kyoto Univ.*, [3]*Grad. Sch. Sci. Eng., Waseda Univ.*)

The structural fluctuations of proteins are important for protein functions. Coarse-grained molecular dynamics (CG-MD) and small angle x-ray scattering (SAXS) are powerful tools to characterize the dynamically fluctuating solution structure. Therefore, we developed a CG-MD-SAXS method that computes SAXS profiles from the structural ensemble obtained by CG-MD.
We applied the CG-MD-SAXS to the canonical, CENP-A, and H2A.B nucleosomes, and we characterized fluctuations of DNA terminal regions in the nucleosomes. In the canonical nucleosome, both two DNA ends were bound to the histone core in the major state, and in the minor state, one DNA end was detached from the histone core. In the CENP-A and H2A.B nucleosomes, both two DNA ends were detached from the histone core.

1Pos076 粗視化シミュレーションによるヘテロクロマチン蛋白質1のヌクレオソームへの結合研究

Heterochromatin protein 1 binding to nucleosomes studied by coarse-grained simulations

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Heterochromatin protein 1 (HP1) binds lysine 9-methylated histone H3 (H3K9me), and plays important roles in forming inactive chromatin structures.
Recent studies suggested that the binding affinity of HP1 to H3K9me-containing nucleosomes is dependent on linker length and nucleosome core-number.
Here, we performed molecular dynamics simulations using a coarse-grained model of HP1 with mono- and di- nucleosomes to analyze their binding.
The simulations revealed the differences in molecular binding dynamics not only between mono- and di- nucleosome but also HP1alpha and its paralogs.

1Pos077 高活性TALE蛋白質の開発とその応用

Development and application of improved TALE protein

Kazuho Ikeda, Yoko Terahara, Kenta Sumiyama, Yasushi Okada (*QBiC, RIKEN*)

Transcription activator-like effector (TALE) is a “designable” DNA binding protein, which is a promising platform for labeling and manipulating of specific genome sequence *in vivo*. One successful example of its application is TALE nuclease, the genome editing enzyme. We recently developed highly active mutant TALEs by introducing amino acid substitutions into specific residues of DNA binding domain and demonstrated TALENs made from this mutant TALE had significantly higher rate of genome editing in zebrafish eggs and mouse embryos. In this study we have applied our mutant TALEs to genome visualizing tools and achieved live imaging of single genomic locus. Further improvement of TALE application is in progress.

1Pos078 粗視化モデルによるダイヌクレオソーム間の相互作用とヒストンテイルの役割

Dinucleosome structure and role of histone tails by coarse-grained model

Hiroo Kenzaki[1], Shoji Takada[2] ([1]*ACCC, Riken*, [2]*Grad. Sch. Sci., Kyoto Univ.*)

Nucleosome is the basic unit of chromatin and assemble to higher-order chromatin structure, and the structure prescribes searching and binding processes of transcription factors. Interactions between nucleosomes are regulated by histone modifications such as acetylation, which change chromatin conformation, however, the mechanism is not elucidated. Thus, we performed molecular dynamics simulation of dinucleosome as for the basic case by coarse-grained protein/DNA model. We simulated dinucleosome dynamics and analyzed structures and interactions between nucleosomes. We also investigate effect of modifications of histone tails by deleting charges of some residues. The role of histone tails and effect of acetylation will be discussed.

1Pos079 G-quadruplex がさまざまの小分子と相互作用することによる構造変化の反応速度論による研究

Kinetics and mechanism of conformational changes by interaction of G-quadruplexes and small molecules

Masato Tanigawa, Takafumi Iwaki (*Fac. Med., Oita Univ.*)

The spectral characteristics, thermostability, and kinetics of the formation of a G-quadruplex were measured in this work using circular dichroism (CD) spectroscopy along with stopped-flow technique. Irinotecan, polyethylene glycol, TMPyP, NMN(Nicotinamide mononucleotide), idarubicin were used for interaction with G-quadruplex as small molecules. Kinetic studies suggest that the small-molecule-induced-folding of G-quadruplex probably proceeds through the different formation of an intermediate associate with K(+). We will report the change of relaxation time, τ, of folding the G-quadruplex in the presence of various small molecules.

1Pos080 蛍光色素から DNA 修飾した単層カーボンナノチューブへのエネルギートランスファーに関する研究

Research on energy transfer from fluorescent dyes to DNA-wrapped single-walled carbon nanotubes

Shusuke Oura, Masahiro Ito, Yoshikazu Homma, Kazuo Umemura (*Tokyo Univ. of Sci.*)

It is known that fluorescent dyes are quenched on single-walled carbon nanotubes (SWNTs). In this study, SWNTs were dispersed with 30-mers of Thymine (T30-SWNTs), and then T30-SWNTs were mixed with fluorescent dye-labeled 30-mers of Adenine or Guanine (T30-SWNTs-FluorA30 or T30-SWNTs-FluorG30). Atomic force microscopy (AFM) showed that both T30-SWNTs-FluorA30 and T30-SWNTs-FluorG30 had similar larger height than T30-SWNTs. Contrarily, according to photoluminescence in a visible light region, quenching effect of the T30-SWNTs-FluorA30 and T30-SWNTs-FluorG30 were 13% and 26% relative to free FluorA30 or FluorG30 at the same concentration, respectively. This is the first effort to examine energy transfer from fluorescent dyes to DNA-pretreated SWNTs.

1Pos081 異なる損傷源が引き起こす DNA 二本鎖切断の DMSO による保護作用の比較

Comparison of the protective effect of DMSO on DNA double-stand break with different injury sources

Masami Noda[1], Yuko Yoshikawa[2], Kenichi Yoshikawa[1], Takahiro Kenmotsu[1], Tadayuki Imanaka[2] ([1]*Doshisha University*, [2]*Ritsmeikan University*)

DMSO (dimethylsulfoxide) is widely and frequently used as a protective agent on living cells against radiations and also freezing, and it is applied for saving cells. We are currently studying double-stand damage of DNA, because of its most serious effect on living cells, including mutation, repairing error and cancer generation. Here, we will report quantitative evaluations on the degree of double-strand damage by difference injury sources, reactive oxygen by light irradiation, gamma ray, and freezing, through the methodology of single DNA observation by fluorescence microscopy. Among the novel discoveries in this study, we may show that minimum threshold concentration of DMSO is necessary for the protective effect, being different from the other injury sources.

1Pos082 超音波パルスによる DNA 切断の促進効果

Pulsing stimuli of ultrasound causes larger damage on DNA than its CW mode: Single DNA observation on double-strand breaks

Rinko Kubota[1], Yusuke Yamashita[1], Yukihiro Kagawa[1], Yuko Yoshikawa[2], Yoshiaki Watanabe[1], Takahiro Kenmotsu[1], Tadayuki Imanaka[2], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life and Medical Sciences, Univ. Doushisha*, [2]*Laboratory of Environmental Biotechnology Research Organization of Science and Technology, Univ. Ritsumeikan*)

Currently, in the medical application of ultrasound, either continuous wave (CW) or pulsed wave has been adapted mostly based on its technical reason. In the present study, we examined the difference of the mode of ultrasonic irradiation to case double-strand breaks (DSBs), by used of single-DNA observation. It was found that pulsed wave of ultrasound caused significantly larger effect on the DSBs its CW mode. In addition, we have evaluated the dose dependence on the DSBs and confirmed that there exist certain threshold and below this threshold there is essentially no damage. Above the threshold, the DSBs increase linearly with the total power of the radiation. We may discuss the mechanism of the ultrasound damage on DNA in relation to the occurrence of percolation.

1Pos083 2D DNA nanostructure to arrange positions of functional molecules

Yuki Matsubara, Akira Suyama (*Univ. of Tokyo, Graduate School of Arts and Sciences*)

The nanotechnology using self-assembly and programmable molecular recognition by DNA hybridization is experiencing great developments especially in the field of DNA nanostructures. These DNA nanostructures become an important way to construct breakthrough materials and functions, because they are hopeful as scaffolds to arrange positions of functional molecules either periodically or aperiodically with a nano-scale precision. In this study, we demonstrated a new 2D periodical DNA nanostructure suitable for aperiodical molecular scaffolds.

1Pos084 エタノールによって引き起こされる DNA の再帰転移現象

Reentrant transition on the higher-order structure of DNA with the increase of alcohol concentration

Yuki Oda[1], Yuko Yoshikawa[2], Tadayuki Imanaka[2], Kingo Takiguchi[3], Masato Hayashi[3], Kenichi Yoshikawa[1] ([1]*Graduate School of Life and Medical Science, Doshisha University*, [2]*Department of Biotechnology, College of Life Sciences, Ritsumeikan University*, [3]*Department of Molecular Biology, Graduate School of Science, Nagoya University*)

We will report that DNA molecules undergo reentrant transition, coil-compact-coil transition, with the increase of ethanol concentration up to 80 v/v%. We found such characteristic transition by ethanol by use of single DNA observation with fluorescence microscopy. In order to study the change of higher-order structure of DNA, we adapted lambda-DNA, 48.5 kbp with the full length of 20 um, because such long DNA molecules behave as a semiflexible polyelectrolyte chain. We have also measured the effect of ethanol on the secondary structure of DNA through CD observation. In the presentation, we will report the comparison on the effect of alcohols with different alkyl moieties for the higher-order structural change of DNA.

1Pos085 Photoactive Yellow Protein に現れる H/D 同位体効果の理論的解析

Theoretical analysis of H/D isotope effect on Photoactive Yellow Protein

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Photoactive Yellow Protein (PYP) is a photosensing protein, whose chromophore, p-coumaric acid (pCA), forms hydrogen bonds (H-bonds) with Tyr42 and Glu46 in the dark state. After comprehensive analyses, it was suggested that a low barrier H-bond (LBHB) would be possible between pCA and Glu46 under the crystallographic condition with vicinal Arg52 deprotonated, while a conventional H-bond would be formed in liquid water where Arg52 was presumably protonated. To validate the above suggestion, we calculated H/D isotope shifts on NMR and UV/Vis spectra of PYP by the combination of ONIOM and multicomponent quantum mechanics (MC_QM); ONIOM(MC_QM:MM), and compared our computational results with the corresponding experimental values.

1Pos086 電解質溶液中におけるマクロアニオンの凝集挙動

Aggregation behavior of macroanions immersed in electrolyte solution

Takuto Sawayama, Ryo Akiyama (*Kyushu Univ., Dept. Chem.*)

To discuss association of acidic proteins we have clarified strong attraction between two macroanions immersed in electrolyte solution by using an integral equation theory with charged hard sphere model. The effective attraction is mediated by cations. Macroanions strongly attract each other only in cation solution whose concentration is about 10-3M, although they repel each other in more dilute or dense electrolyte solution. Phase behaviors for the system are studied by using Monte Carlo simulation on the basis of the effective interaction. The aggregation behavior depends on not only salt concentration but also protein concentration. Taking account of cations adsorption on the macroanions, we can explain the related experimental result.

1Pos087 演題取り消し

1Pos088 シミュレーション・データマイニングによるリガンド結合系における水分子の振る舞い

Simulation-based data-mining approach for water behavior of ligand-binding system

Taku Mizukami[1], Viet Cuong Nguyen[3], Tu Bao Ho[2], Hieu Chi Dam[2] ([1]*Sch. of Materials Sci., JAIST*, [2]*Sch. of Knowledge Sci., JAIST*, [3]*HPC Systems Inc.*)

Water plays an important role in biological systems. Biomolecules express their functions whose mechanisms are strongly influenced by interactions with water. Because of the strong hydrogen bonding and the fluctuations, the nature of water behavior remains a hard task to clarify.
To overcome the difficulty, we applied a data-driven approach to the water dynamics unifying molecular dynamics simulations and data-mining techniques. The mining on the water behavior in protein solution resulted in several classes. Reproduced physicochemical features showed that a single class of water behavior corresponded to the protein hydration water. Other water classes indicate a variety of water dynamics.
In the presentation, we will show the mining results on the ligand-binding system.

1Pos089 Cell differentiation and Reprogramming: A minimal model

Ashwin S.S., Masaki Sasai (*Nagoya University, Japan*)

Using the epigenetic landscape perspective, differentiation/reprogramming can be regarded as transitions between basins corresponding to pluripotent and differentiated cell states. Differentiatiion is thought to be induced via stochastic fluctuations in the chromatin and other cellular processes on the other hand reprogramming is induced by a driving mechanism provided by the so called Yamanka factors. We provide a theoretical construction of a minimal model, whose landscape exhibits differentiation and reprogramming dynamics. Our model exhibits a prominent intermediate state during the reprogramming, which is less significant during differentiation, consistent with experiments.

1Pos090 多能性幹細胞の分化における１細胞解析

Single-cell-based analysis of differentiation of pluripotent stem cells

Ayumu Kano[1], Yuta Yamamoto[1], Shogo Nakamura[1], Atsushi Maruyama[1], Olga M De Sousa[2], Masahiro Iwahashi[3], Toshiyuki Akaike[4], Kiyoshi Ohnuma[1] ([1]*Nagaoka University of Technology Bioengineering*, [2]*Faculty of Engineering, Science and Technology National University of East Timor*, [3]*Nagaoka University of Technology Electronics and Information Engineering*, [4]*Tokyo Institute of Technology, Yokohama, Japan 226-8501*)

Embryonic stem (ES) cells and induced pluripotent stem (iPS) cells are a good model for studying early development of mammalian in vitro. We developed a system to trace a single-cell of mouse ES (mES) cells and human iPS (hiPS) cells. Comparison of daughter mES cells derived from common mother mES cells showed the correlation between the daughter cells, suggesting the daughter cells keep memory of mother cells. Moreover, deviation of both division time and GFP strength of mES cells in differentiation condition were larger than that in undifferentiation condition, suggesting the responses to condition change is uneven. Single-cell level observation of ES/iPS cells will deepen our understanding of developmental stability for future application in reproductive medicine.

1Pos091 植物の中の局所微小管と細胞成長の関係について
On the Relationship between Local Microtubule and Cell Growth in Plants

Satoru Tsugawa, Chun-Biu Li, Tamiki Komatsuzaki (*Research Institute for Electronic Science*)

A longstanding question in developmental biology is how organs reach their final shapes despite their cells having high variability in shape and size at the microscopic level. To address this issue, the behavior of the cortical microtubules is receiving a lot of attention in plants because they are thought to play an essential role in morphogenesis. We suggest an quantitative assessment for local microtubule and discuss the relationship between the local microtubule and heterogeneous growth in plants. Interestingly, we found that the microtubule fluctuation is anti-correlated with the maximal length difference of cells at the subcellular level, neighborhood level, and organ level.

1Pos092 Control of Morphological Dynamics of Myoblasts Using Stimulus-Responsive Hydrogels Cross-linked by Host-Guest Interactions

Marcel Hoerning[1], Masaki Nakahata[2], Akihisa Yamamoto[1], Yoshinori Takashima[2], Akira Harada[2], Motomu Tanaka[1] ([1]*iCeMS, Kyoto University*, [2]*Osaka University*)

Polymer substrates with tunable elasticity have widely been used to regulate single cells [Engler, et al. Cell 126, 2006] and multicellular tissues [Hoerning, et al. Biophys. J. 102, 2012]. As a more biologically relevant model of dynamic micro-environments, stimulus-responsive materials that can reversibly alter the elasticity have been developed [Yoshikawa, et al, JACS, 133 2011]. In this study, we apply a new class of hydrogels cross-linked by supramolecular host-guest interactions [Nakahata, et al, Nature Commun. 2, 2011]. After systematic optimization of the system, the morphological dynamics of C2C12 myoblast in response to changes in substrate stiffness was monitored in real time.

1Pos093 CryoEM structure of muscle thin filament with the tropomyosin and troponin complex

Yurika Yamada[1], Keiichi Namba[1,2], Takashi Fujii[1,3] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*QBiC, RIKEN*, [3]*JST PRESTO*)

Muscle contraction is driven by cyclic interactions of myosin in the thick filament with thin filament composed of actin, tropomyosin (Tm) and troponin (TnC, TnI, TnT). It is thought that the binding of Ca2+ released from sarcoplasmic reticulum to TnC causes a conformational change of Tm on the actin filament to allow actin-myosin interaction. To understand this regulatory mechanism, it is necessary to elucidate the structure of thin filament at high resolution. We developed a method to purify intact, Ca2+-free and Ca2+-bound thin filaments from skeletal muscle of a crab, Portunus trituberculatus, at high yield. A cryoEM density map of thin filament at around 20 Å resolution in the absence of Ca2+ shows interesting features of actin-Tm-Tn interactions never seen before.

1Pos094 ナノイメージングによるマウス心臓の *in vivo* サルコメア動態解析
High-speed live imaging of single sarcomere dynamics in the beating mouse heart *in vivo*

Fuyu Kobirumaki[1], Kotaro Oyama[1,2], Togo Shimozawa[4], Seine Shintani[5], Erisa Hirokawa[1], Takako Terui[3], Shin'ichi Ishiwata[2], Norio Fukuda[1] ([1]*Dept. Cell Physol., Jikei Univ. Sch. Med.*, [2]*Dept. Physics, Waseda Univ.*, [3]*Dept. Anesth., Jikei Univ. Sch. Med.*, [4]*Dept. Life Sci. Med. Biosci., Waseda Univ.*, [5]*Dept. Physics, Univ. Tokyo*)

To explore the molecular mechanisms of cardiac muscle contraction under physiologic conditions, we directly imaged the motions of a single sarcomere in the beating heart *in vivo*. In the present study, we developed a high-speed (100 fps) high-resolution (20 nm) imaging system for myocardial sarcomeres in living mice. We found that (1) the working range of sarcomere length (1.90 and 1.68 μm in diastole and systole, respectively) existed on the shorter resting distribution side and (2) the left ventricular developed pressure was linearly correlated with the sarcomere length change between diastole and systole on the order of 100 nm. The present findings provide the first direct evidence for the tight coupling of sarcomere dynamics and ventricular pump functions.

1Pos095 F-アクチンの水和状態の円二色性分光と DSC 測定による検討
Study on the correlation of hydration state, ternary structure and heat capacity of F-actin

Takaya Yamaguchi, Ryotaro Chisima, George Mogami, Makoto Suzuki (*Graduate School of Engineering, Tohoku University*)

Hydration shell of F-actin was previously found to be composed of restrained water with dielectric relaxation (DR) frequency f_2 lower than that of bulk f_w and hypermobile water (HMW) with DR frequency $f_1 > f_w$. (Kabir et al., Biophys. J. 2003, 85, 3154-3161) According to recent structural studies by Fujii et al.(2010) and Oda et al.(2010), there appeared clear difference between Mg- and Ca-bound F-actins. In this study, we measured the CD spectra from 270 to 300 nm to examine the ternary structure of F-actin and the heat capacity of F-actin solution in the Mg- and Ca-bound states by DSC. As a result, we found that the ternary structure of F-actin was loosened in Ca-F-actin and F-actin with more HMW had lower heat capacity.

1Pos096 電子顕微鏡によるフォルミン mDia1 のアクチンフィラメント端結合構造の解析
Structural analysis of binding state of formin/mDia1 to actin filament by electron microscopy

Mizuki Matsuzaki, Akihiro Narita (*Grad. Sch. Sci., Nagoya Univ.*)

Formin is potent regulator of actin dynamics. With effects on a filament nucleation rate, filament elongation rate and barbed-end capping protein function, the net activity of formins is to control actin filament assembly. We investigated suitable conditions for structure analysis of binding states of FH2 domain of mDia1 (one of mouse formin) to the actin filament by electron microscopy. We found a condition under which mDia1 without aggregation has high activity. The obtained actin filaments bound to mDia1 have enough quality to analyze structure by electron microscopy. Now we started the structure analysis of mDia1 binding to actin filament.
We also observed the actin filament by using AFM, STEM and freeze replica technique.

1Pos097 アクチン繊維の骨格形状と滑り運動時に生じる破断の原因
A cause for breaking of sliding actin filament on myosin

Katsunari Sakamoto, Shigeru Sakurazawa (*Future Univ. Hakodate*)

It is known that sliding velocity of actin filaments depends on ATP concentration. However, it is tend to decrease at the specific ATP concentration area. In addition, sliding actin filament is broken occasionally. As the first step toward figuring out energy transduction system of muscle protein, we investigated the cause of breaking of an actin filament. Firstly, we observed the rate of break at various ATP concentration. Also, we searched the curvature before and after actin is broken. As a result, the rate of break is the highest at the point which sliding velocity is decreased. The curvature before break was large and the filament was broken from the center of large curvature. We suggest that actin filament was broken as a means of releasing energy.

1Pos098 Probing stator-protein dynamics of bacterial flagellar motors

Chien-Jung Lo, Tsaishun Lin (*Department of Physics, National Central University, Taiwan*)

The bacterial flagellar motor is a natural electrical rotary molecular machine. The stator-unit consists of MotA/B proteins couples ion-flux to motor rotations. More than 10 stator-units in a motor turn over dynamically in response to the cellular energetical conditions and external loads. However, spatial features and temporal dynamics of stator-units have never been studied thoroughly. We have built a super-resolution fluorescent microscope with 10 nm resolution to study stator-units dynamics using dual color labeling on rotor- and stator- proteins. We will present protein counting, spatial distribution and dynamical properties of stator proteins.

We thank Seiji Kojima, Hajime Fukuoka, Akihiko Ishijima and Michio Homma for the bacterial strains.

1Pos099 Effect of Microtubule Deformation on Kinesin-based Cargo Transportation In Vitro

Tanjina Afrin[1], Arif Md. Rashedul Kabir[2], Daisuke Inoue[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

Biomolecular motor protein and microtubule (MT) together ensure the temporal and spatial demands for intracellular transport of cargoes. In some cases, MT rails are found to be highly deformed in cells and may interrupt the intracellular cargo transportation. The interrupted cargo transportation due to track deformation has been suspected to be linked to many neurodegenerative diseases in particular cargo transport defect in axons. However, the effect of the deformed MT track on the kinesin-based cargo transportation is yet to be understood. We investigated the kinesin-based cargo transportation along the buckled MT track in vitro and observed that the cargo transportation was decelerated along the buckled MT track depending on the extent of buckling of the MT track.

1Pos100 High resolution structural analysis of the flagellar hook of Salmonella

Peter Horvath[1], Tomoko Miyata[1], Hiroko Takazaki[1], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Riken QBiC*)

Equipments and techniques for CryoEM methods for structural studies of macromolecules have rapidly advanced in recent years. These developments allow near-atomic resolution structure determination, which is inevitable for understanding underlying biological mechanisms. The bacterial flagellar hook functions as a universal joint. The highest resolution structure available so far is 7.1 Å, and this was achieved with a CCD camera. To gain more detailed information about the flexible mechanism of the hook, we are now optimizing the conditions for data collection using a direct electron detector and the use of software based on multivariate statistical analysis.

1Pos101 マルチスケール MD による V1ATPase の回転機構の解明
Rotation mechanism of V1-ATPase studied by multi-scale MD simulation

Yuta Isaka[1], Yuuichi Kokabu[1], Toru Ekimoto[1], Takeshi Murata[2,3], Mitsunori Ikeguchi[1] ([1]*Grad. Sch. of Med Life Sci., Yokohama City Univ.*, [2]*Fac. of Sci., Chiba Univ.*, [3]*JST, PRESTO*)

V-ATPase is a molecular-motor acting as a proton pump in cell membranes.Recently, high-resolution crystal structures of *Enterococcus hirae.* V_1-ATPase, which is a soluble domain of V-ATPase, were determined.In the structure, V_1-ATPase adopts the different conformations coupled with ATP hydrolysis, and the conformational changes may induce the rotation of the central stalk.

To elucidate the relationship between the conformational changes and the stalk rotation, we performed 100 ns all-atom (AA) MD simulations in the three states of V_1-ATPase. Then, we linked the states using the course-grained (CG) MD simulations consistent with AA-MD fluctuations, indicating the transition pathway of the conformational changes.

1Pos102 アクチン-ミオシン結合における水の役割の理論解析
A theoretical analysis on water roles in actin-myosin binding

Hiraku Oshima, Tomohiko Hayashi, Masahiro Kinoshita (*Inst. Adv. Energ., Kyoto Univ.*)

Although the binding of actin and myosin has intensively been studied, its molecular mechanism, in particular the effect of surrounding water molecules, is still not clear. Here we investigate the water roles in the actin-myosin binding. We calculate changes in thermodynamic quantities upon the binding of F-actin and myosin subfragment-1 using a statistical-mechanical approach combined with molecular models for water. The changes are decomposed into physically insightful components. We find that the water-entropy gain plays an important role in the binding. It is experimentally known that the binding power becomes weaker at low temperatures. The temperature dependence of the hydration effects is also discussed.

1Pos103 タンデム型エフェクターによる T3SA 内エフェクター分泌機序の評価

Analysis of Type 3 secretion mechanism by "tandem-connected effectors"

Kyoko Momiyama, Takashi Ohgita, Kentaro Kogure (*Kyoto Pharm. Univ.*)

Type 3 secretion apparatus (T3SA) is a needle-like molecular machine and it is used for direct injection of effector proteins helping bacterial pathogenicity. Now, however, how effector goes out through T3SA is unclear because effector is too small to be observed under microscope. To clarify this, we construct an evaluation system for effector motion by using tandem-connected effectors. By using this system, if bacteria secrete effectors at constant speed, the secretion time should become longer in proportion to the length of proteins. Therefore we could know about secretion speed of effectors. In addition, the tandem of varieties of effector may effect some change in secretion. Now we continue experiments to build the system.

1Pos104 蛍光性 NTA-Ni の合成とフォトクロミック分子で修飾した有志分裂キネシン Eg5 への蛍光ラベリング

Synthesis of fluorescent NTA-Ni and its application to the fluorescent labeling of mitotic kinesin Eg5 modified with photochromic molecule

Yuki Tamura, Kei Sadakane, Ryoma Yamamoto, Kentaro Saito, Shinsaku Maruta (*Division of Bioinformatics, Graduate School of Engineering, Soka University*)

We have been trying to control the function of Mitotic kinesin Eg5 by using photochromic molecules. To modify Eg5 with photochromic molecules, we have prepared the Eg5 mutants which have a single reactive cysteine. However, the mutants which cysteine residue are occupied with the photochromic molecules failed to be modified with thiol reactive fluorescent probe. In this study, we utilized fluorescent probe bound NTA-Ni to label the His tagged Eg5 modified with photochromic molecules. According to the methods of Soh et al., Dansyl-NTA and other fluorescent NTA were synthesized and conjugated with Ni2+. The Dansyl-NTA-Ni2+ bound to His tagged Eg5. Using the fluorescent NTA-Ni2+, interaction of photo-regulated Eg5 with microtubules was monitored.

1Pos105 構成論的手法による真核生物鞭毛運動の機能再構築

Reconstruction of flagellar motility by the bottom-up strategy

Junya Kirima[1], Misaki Shiraga[2], Hiroaki Kojima[3], **Kazuhiro Oiwa**[1,3,4] ([1]*Grad. Sch. Univ. Hyogo*, [2]*F. Sci., Univ. Hyogo*, [3]*NICT, Adv. ICT. Res. Inst.*, [4]*CREST, Biodynamics*)

The complexity of the flagellar axoneme is derived from modular building blocks assembled hierarchically. To find clues of this complexity, using flagella of *Chlamydomonas* as the model, we have performed in vitro reconstitution of axonemal structures and functions in bottom-up manner. We reconstituted crude outer arms extracted with high salt from wild-type axonemes into the outer-armless (*oda1*) mutant axonemes. This reconstitution recovers the frequency of the ciliary beating at low Ca^{2+} and shows waveform switching at high Ca^{2+}. Furthermore, we confirmed the addition of the crude extract forms the bundle of microtubules polymerized on the axoneme fragments. Linear arrays with 24 nm structural repeat on microtubules were found as previously reported.

1Pos106 アゾベンゼン誘導体を利用したキネシン二量体化の光制御

Photo-regulation of kinesin dimerization using bifunctional azobenzene derivatives

Haruka Fujio[1], Yasunobu Sugimoto[2], Kazunori Kondo[3], Shinsaku Maruta[1,3] ([1]*Div. of Bioinfo, Gra. Sch. of Eng., Univ. of Soka*, [2]*Nagoya Univ., SR Research Center*, [3]*Dep. of Bioinfo., Fac. of Eng., Univ. of Soka*)

The aim of this study is to control dimerization and motor activity of kinesin utilizing the photo-reversible conformational change at the coiled-coil stalk region induced by photochromic molecules. We employed two bifunctional azobenzene derivatives which crosslinking spans are altered significantly by cis-trans photo-isomerization of azobenzene moiety upon ultraviolet and visible light irradiations reversibly. We also prepared the kinesin mutants which have two reactive cysteine residues at the coiled-coil stalk region. The kinesin mutants were intra-molecularly cross-linked by the bifunctional azobenzene derivatives. X-ray scattering experiments revealed that the cross-linked kinesin changes its conformation upon ultraviolet and visible light irradiations.

1Pos107 ATP 合成酵素の回転運動の幾何学的位相モデル

Geometric-phase model for the rotary motion of ATP synthase

Tomohiro Yanao (*Grad. Sch. Sci. & Eng., Waseda Univ.*)

This study proposes a geometric model for the rotary motion of the central shaft of FoF1-ATPase. The model is highly coarse-grained, consisting of five mass points connected with four elastic springs. This model utilizes a geometric phase arising from a cyclic motion of dihedral angles to achieve a stepwise rotary motion by 120 degrees without generating total angular momentum. Such geometric phase is analogous to the one that arises in the so-called "falling cat" phenomenon, where a falling cat changes its overall orientation by changing its shape under conditions of zero total angular momentum. We place this geometric model in a thermal environment and compare its functional efficiency with that of a conventional rigid-rotor model in terms of energetics.

1Pos108 *Mycoplasma mobile* の滑走に必須な Gli123 タンパク質の構造解析

Structure analyses of Gli123 protein, essential for gliding of *Mycoplasma mobile*

Daiki Matsuike, Yuhei O. Tahara, Tasuku Hamaguchi, Makoto Miyata (*Grad. Sch. Sci., Osaka City Univ.*)

M. mobile, a fish pathogen glides on solid surfaces by a unique mechanism. Three large proteins clustering on the surface of the gliding machinery are essential for this mechanism. In this study, we focus on the structure of Gli123, a 123-KDa protein responsible for the assembly of surface gliding proteins. The Gli123 protein and its recombinant were isolated from *M. mobile* and *Escherichia coli* cells, respectively. Gel filtration chromatography showed that both of them form a homodimer. Negative - staining electron microscopy followed by single particle analyses from 13,000 particle images showed a three-dimensional molecular shape like a "mushroom", with dimensions, 20.0, 14.5, and 16.0 nm. Detailed structural analyses are now undertaken.

1Pos109 カタユウレイボヤ精子鞭毛における内腕ダイニンのキャラクタリゼーション

Characterization of inner arm dyneins from sperm flagella in *Ciona intestinalis*

Osamu Kutomi[1], Katsutoshi Mizuno[1,2], Keiko Hirose[3], Lixy Yamada[4], Hitoshi Sawada[4], Kogiku Shiba[1], Kazuo Inaba[1] ([1]*Shimoda Mar. Res. Ctr.*, [2]*Dartmouth College*, [3]*Biomed. Res. Inst., AIST*, [4]*Sugashima Marine Biological Lab., Grad. Sch. Sci., Nagoya Univ.*)

Inner arm dyneins (IADs) of cilia and flagella are classified into several subclasses. Each subclass is thought to play distinct roles in motility, but little is known about the IADs from metazoan sperm flagella. Here we isolated two-headed IAD (dynein-f) and several single-headed IADs from sperm flagella of ascidian *Ciona intestinalis*. Although their molecular compositions and structures studied by mass spectrometry and electron microscopic analysis were similar to those of *Chlamydomonas* IADs, we found a novel 32-kDa axonemal protein as a subunit of dynein-f. This subunit contains a BLUF domain (sensors of blue-light using FAD), and we named this protein as DYBLUP (Dynein-associated BLUF Protein). Biochemical and structural features of *Ciona* IADs will be discussed.

1Pos110 Dynactin is a biphasic regulator to promote and inhibit dynein motility

Takuya Kobayashi[1], Takuya Miyashita[1], Hatsuha Kajita[1], Kei Saito[1], Takashi Murayama[2], Yoko Y. Toyoshima[1] ([1]*Grad. Sch. Arts and Sci., Univ. of Tokyo*, [2]*Sch. Med., Juntendo Univ.*)

p150 subunit of dynactin has microtubule binding and dynein binding regions. There are two splicing variants, DCTN1A and DCTN1B, differing in K-rich region relating to microtubule binding affinity. Recent reports indicated that the dynein-dynactin-BICD2 complex caused the super-processive movement. We also obtained the similar motility with DCTN1A. However, DCTN1B did not induce the super-processive movement of the complex but dissociated it from the microtubules. The dissociating effect was caused by CC1 region of p150 and this effect was partly observed with DCTN1A. Thus, dynactin has two regions which induce the opposing effects, microtubule binding and dissociation, in a p150 subunit.

1Pos111 カビ由来セルラーゼ Trichoderma reesei Cel6A および Cel7A の逐次運動の１分子蛍光観察

Single-molecule fluorescence imaging analysis of processive movement of fungal cellulases Trichoderma reesei Cel6A and Cel7A

Tomoyuki Tasaki[1], Akihiko Nakamura[2,3], Daiki Ishiwata[2,3], Hiroyuki Noji[1], Ryota Iino[2,3] ([1]*School of Engineering, The University of Tokyo*, [2]*Okazaki Inst. Integ. Biosci., NINS*, [3]*School of Physical Sciences, SOKENDAI*)

High-speed AFM (HS-AFM) previously verified that a fungal cellulase Trichoderma reesei Cel7A (TrCel7A) is a linear molecular motor which processively hydrolyzes and moves on crystalline cellulose. However, the movement of a related enzyme, Trichoderma reesei Cel6A (TrCel6A) has not been observed with HS-AFM. Since TrCel6A has more “opened” loop structure around catalytic site compared with that of TrCel7A, there is a possibility that tapping force of HS-AFM destabilizes TrCel6A-cellulose interaction and inhibits TrCel6A movement. With single-molecule fluorescence imaging, here we show that both TrCel7A and TrCel6A move unidirectionally on crystalline cellulose with velocities of ~5 nm/s and ~8 nm/s, respectively. Therefore, TrCel6A is also a linear molecular motor.

1Pos112 High-speed angle-resolved imaging of domain motion of the catalytic β subunit of F_1-ATPase

Sawako Enoki[1], Ryota Iino[2], Yoshihiro Minagawa[1], Yamato Niitani[3], Michio Tomishige[3], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem, Grad. Sch. Eng.*, [2]*Okazaki Inst. Integ. BioSci., NINS*, [3]*Dept. Appl. Phys, Grad. Sch. Eng.*)

F_1-ATPase is an ATP-driven rotary molecular motor. Rotation of the rotor γ subunit is driven by coordinated conformational change of three catalytic β subunits in the stator ring. To elucidate the mechanism how each catalytic β subunit rotates the γ subunit, we observed conformational change of catalytic β subunit by monitoring orientation of gold nanorod with dark-field imaging at microsecond temporal resolution. As a result, we could resolve reversible changes of the catalytic β subunit between open and closed states driven by ATP hydrolysis reaction. Furthermore, we found the half-closed conformation probably caused by temperature sensitive reaction and estimated the force generated by β subunit during conformational change between open and closed conformation.

1Pos113 微小管モータータンパク質による微小管構造の識別機構の解析

In vitro analysis of smart motor hypothesis

Tomohiro Shima[1,2], Yasushi Okada[2] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*QBiC, Riken*)

Kinesin and dynein transport a wide variety of cellular cargoes along microtubules. It is still unclear how these cargoes reach correct destinations. Mainly based on recent neuron studies, it has been proposed that microtubules can take several conformations and subsets of kinesin distinguish such structural difference of microtubules, so that the kinesins transport their cargoes along correct microtubules, leading to the correct destinations. As the first in vitro evidence for the “smart motor hypothesis”, we have shown that conventional kinesin distinguishes difference between GTP-type and GDP-type microtubules. Here, we show the details of interaction between the microtubules and kinesin, and discuss its physiological significance.

1Pos114 バクテリアべん毛モーターにおける二軌道回転

Two orbital rotation of bacterial flagellar motor

Yoichiro Sawano[1], Yuichi Inoue[1], Akihiko Ishijima[2] ([1]*IMRAM, Tohoku University*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

Torque generation of bacterial flagellar motor has been clarified assuming that the rotation occurs in only one orbit (Chen and Berg, 2000). In the bead-rotation assay, however, we found the rotation could consist of multiple orbits. Here we analyzed the multiple orbits in the rotation using asymmetric beads to understand the mechanism of the motor rotation. A small fluorescent bead was combined with a large non-fluorescent bead to observe rotation at the stub of a truncated flagellum. Bright field images showed the slow rotation of ~10 Hz, whereas fluorescence images showed the fast rotation of ~80 Hz, suggesting the two independent orbits. Analysis of rotational speed and torque in each orbit will be presented to discuss why rotation occurs in two orbits.

1Pos115 F_1-ATPase の回転拡散係数に関する理論研究

Theoretical study of rotational diffusion for F_1-ATPase

Ryota Shinagawa, Kazuo Sasaki (*Dept. Applied Physics, Tohoku Univ.*)

F_1-ATPase (F_1) is a motor protein which rotates its γ-shaft (rotor) using the free-energy released by the hydrolysis of adenosine triphosphate (ATP). The efficiency of conversion of this free-energy into F_1's mechanical motion reaches roughly 100%. A recent measurement of rotational diffusion coefficient (D) of F_1 without ATP suggests that we can obtain the insight into the rotor-stator interaction of F_1 from the dependence of D on the external torque (R. Hayashi *et al.*, Phys. Rev. Lett., to be published). We calculate theoretically D of F_1 in the presence of ATP, using a model similar to the one proposed by K. Kawaguchi *et al.*, Biophys. J. (2014). Then, we discuss what kind of information on the rotor-stator interaction can be obtained.

1Pos116 べん毛モータートルク発生に関わる構成蛋白質 FliG の多量体の検出

Identification of multimeric forms of FliG, a flagellar motor component for torque generation

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The rotation of the *Escherichia coli* flagellar motor is driven by proton-motive force. Its torque is generated by an interaction between the stator component MotA and the rotor component FliG. This interaction must be modulated upon switching in rotational direction (counterclockwise/clockwise) in response to chemotactic signals. FliG forms a well defined ring structure, but its biogenesis and conformational changes are largely unknown. In this study, we characterized FliG multimers using native PAGE. Western blotting analyses indicated that they were a monomer, a dimer, a tetramer and a hexamer of FliG. We are currently examining whether a mutant FliG protein with fixed rotational direction makes multimers with wild-type or different mutant FliG proteins.

1Pos117 非平衡効果が引き起こす F_1 分子モーターの非アレニウス型温度依存性

Non-Arrhenius Type Temperature Dependence of F_1 Molecular Motor Induced by Non-equilibrium Effects

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F_1-ATPase(F_1) is a rotary motor driven by ATP hydrolysis, whose chemo-mechanical coupling has been extensively studied. Here, we study the temperature dependence of F_1 to unveil the role of thermal fluctuations. Combining single F_1 observation and time series analysis, we model the F_1 catalytic reactions in terms of a reaction-diffusion formalism where the catalytic power is modulated by the rotary angle fluctuations. We found that the temperature affects both the fluctuation amplitude and the angle correlation. Specifically, non-equilibrium effects can arise when time scales of reaction and diffusion are comparable, leading to non-Arrhenius behaviors. The biophysical implications will be also discussed in this talk.

1Pos118 走化性シグナル CheY-P を結合したべん毛基部体スイッチ複合体の構造

Structure of the flagellar basal body swich complex with chemotaxis signal protein CheY-P

Tomoko Miyata[1,2], Takayuki Kato[2], Yusuke V. Morimoto[1,2], Hideyuki Matsunami[3], Keiichi Namba[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Trans-Membrane Trafficking Unit, OIST*)

Many bacteria swim by flagella rotating bidirectionally. The three switch proteins, FliG, FliM and FliN, form the C-ring on the cytoplasmic face of the MS ring and control counterclockwise-clockwise (CCW/CW) switching of the motor rotation. CheY is a response regulator in bacterial chemotaxis, and phosphorylated CheY (CheY-P) binds to FliM/N and changes the rotational direction. We previously reported the C ring structures locked in CCW (che deletion strain) and CW (FliG-ΔPAA strain). Comparison of the two strucures showed differences in the position of the C ring and the subunit arrangement in its outer wall. We will now report *in vitro* reconstitution and the structure of the CheY-P bound C ring and discuss the switching mecanism of flagellar rotation.

1Pos119 好熱菌 F_1 の βE190 残基は構造を安定化することにより、F_1 の高効率エネルギー変換に貢献する

TF_1 βE190 residue contributes to high energy transduction efficiency by stabilizing the structure of the enzyme

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F_1-ATPase is a motor protein driven by ATP hydrolysis. It converts chemical free energy change into the kinetic energy of the rotation. Previously, Toyabe found that the maximum work of wild type TF_1 estimated from stall torque is nearly equal to the free energy change of ATP hydrolysis, and reached the conclusion that TF_1 has very high energy transduction efficiency (PANS 2011). In order to find out the mechanism of this high efficiency, we compared the response to external torque of wild type with that to βE190D mutant. In the last annual meeting, we reported that this mutant showed smaller stall torque than wild type. Here we show that the mutant cannot form stable $\alpha_3\beta_3$ complex, inferring this instability is a cause for the low energy conversion efficiency.

1Pos120 2 種類のイオンで駆動する細菌べん毛モーターのエネルギー変換機構の解明

Analysis of bacterial flagellar rotation driven by dual ion

Kenta Arai[1], Yuka Takahashi[2], Masahiro Ito[2], Yoshiyuki Sowa[1] ([1]*Hosei Univ.*, [2]*Toyo Univ.*)

The bacterial flagellar motor rotates at high speed by converting the free energy gained from the ion current across the membrane. The motor consists of a rotor and multiple stator complexes. Coupling ions are determined by the characteristics of the stator complex, which acts as an ion channel. For example, Escherichia coli has a proton-driven motor with MotAMotB stator. Recently, it was reported that MotPMotS stator of Bacillus alcalophilus, which acts as sodium- and potassium ion channel, can interact with a rotor of E. coli flagellar motor.

In this study, to investigate the energy conversion mechanism that is common to the bacteria flagellar motor, we measured the motor output of MotPMotS motor in E. coli under controlled sodium- and potassium-ion motive force.

1Pos121 The Growth Rate of Vibrio alginolyticus Polar Flagellum Decays Exponentially with Flagellar Length

Mei-Ting Chen (*National Central University*)

Bacterial flagella are self-assembled external tubular filaments. Flagellins, flagellum monomers, are pumped out from the basal body associated with six integral membrane proteins and three soluble proteins through the nanometer transportation system. As flagellin reaches the end of flagellar filament, it becomes the new extending part of the filament. In order to study this nanometer size self-assembled system, we have developed a fast flagellar protein binding assay to directly monitor the length of the polar flagella of Vibrio alginolyticus. It enabled us to track a single flagellum length within few minutes. We measured the growth rate at 30 and 37 oC. We found that the filament growth rate decrease exponentially with filament length at both temperatures.

1Pos122 マスト細胞の脱顆粒におけるミトコンドリアカルシウムユニポーターの役割
Role of mitochondrial calcium uniporter in mast cell degranulation

Tadahide Furuno, Narumi Shinkai, Masanari Ishikawa, Yoshikazu Inoh, Mamoru Nakanishi (*Sch. Pharm., Aichi Gakuin Univ.*)

The Ca^{2+} uptake into the mitochondrial matrix influences ATP production, Ca^{2+} homeostasis, and apoptosis regulation. We previously reported that mitochondrial Ca^{2+} uptake in two steps played a role in mast cell function through cytoplasmic Ca^{2+} buffering. Here we have analyzed how impaired expression of mitochondrial Ca^{2+} uniporter (MCU) and its regulatory component mitochondrial Ca^{2+} uptake 1 (MICU1) affects mast cell degranulation. Both mitochondrial Ca^{2+} uptake and degranulation after antigen stimulation were significantly suppressed in MCU-knockdown (KD) cells but not in MICU1-KD cells. This suggested that MCU regulated the mast cell response via mitochondrial Ca^{2+} uptake.

1Pos123 1 分子観察によるガングリオシドのダイマー形成機構の解明
Unraveling of mechanisms of ganglioside dimer formation as revealed by single-molecule imaging

Kenichi Suzuki[1], Hiromune Ando[1,2], Naoko Komura[1,2], Ayano Yamazaki[2], Hideharu Ishida[2], Koichi Furukawa[3], Kenichi Morigaki[4], Akihiro Kusumi[1], Makoto Kiso[1,2] ([1]*iCeMS, Kyoto Univ.*, [2]*Dpt. Appl. Biol. Sci., Gifu Univ.*, [3]*Dpt. Biochem., Nagoya Univ.*, [4]*Res. Ctr. Env. Genomics, Kobe Univ.*)

Gangliosides are considered to strongly interact with rafts in cell plasma membranes. However, how they behave in rafts have been unknown due to the lack of their fluorescent analogues. Here, we synthesized 7 kinds of fluorescent ganglioside probes that behave like the endogenous molecules. Surprisingly, single-molecule observation revealed that all the fluorescent ganglioside probes transiently formed homodimers. Furthermore, we determined which functional groups in sugar-chains are important for the homodimer formation by the observation with structurally modified analogues of gangliosides. We propose a new raft concept that specific sugar-chain interactions induce ganglioside homodimer rafts which are likely one of the basic unit for raft organization and function.

1Pos124 ミトコンドリア輸送・ATP 産生の同時測定と相関評価
Simultaneous imaging and correlation analysis of mitochondrial trafficking and its ATP production

Rika Suzuki, Kohji Hotta, Kotaro Oka (*Keio University, BioPhysics and NeuroInformatics Lab.*)

Mitochondrial transport towards high-energy-demand places such as spines and growth cones (GC) suggests mitochondrial ATP production are related to its motility, no research has measured them directly and simultaneously. In this study, we quantified mitochondrial ATP levels and mitochondrial dynamics simultaneously in neurons under physiological conditions. About 70 percent of mitochondria were immobile, while the rest 30 percent moved to either growth cones or cell bodies. Detailed image processing and correlation analysis revealed that ATP levels of mitochondria moving to GC have negative correlation with its velocity. This finding gives advantageous for understanding the relationships between mitochondrial energy status and its dynamics.

1Pos125 好中球様 HL-60 細胞の繰返し基質伸展刺激下での運動方向決定
Directional migration of neutrophil-like HL-60 cells by cyclic substratum stretching

Chika Okimura[1], Kazuki Ueda[1], Yuichi Sakumura[2,3], Yoshiaki Iwadate[1] ([1]*Facult., Sci., Yamaguchi Univ.*, [2]*Sch. Info. Sci. Tech., Aichi Pref. Univ.*, [3]*Grad. Sch. Biol. Sci., Nara Inst. Sci. Tech.*)

Crawling cells respond to mechanical stimuli from the substrata to decide their migration properties. For example, cyclic stretching (CS) of elastic substrata makes Dictyostelium cells migrate in a direction perpendicular to the stretching via a myosin II-related process. Here, we applied CS stimuli to neutrophil-like differentiated HL-60 cells, which localize myosin IIA equally on both stretching sides in the cell and migrate in a direction perpendicular to the stretching. High probability of a switch of migration direction to that perpendicular to the stretching was the main cause of such directional migration. This directional migration is not limited to Dictyostelium cells but is likely to be common in other fast-crawling cell types such as HL-60 cells.

1Pos126 構造的微小管結合蛋白質を介した微小管とアクチンフィラメント間の相互作用
Interaction between microtubules and actin filaments via structural microtubule-associated proteins

Miyuki Shiga[1], Shouma Saitou[1], Yurika Hashi[1], Kazushi Matui[1], Susumu Kotani[2], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Sustain. Environ. Eng., Muroran Inst.*, [2]*Kanagawa University*)

Structural microtubule-associated proteins, MAP2, MAP4, and tau, have microtubule-binding and assembly-promoting activities. Although we revealed that microtubule-binding domain of MAP4 could bind to actin filaments (F-actin) the physiological function remains unknown. Microtubule-F-actin interaction plays important roles in various cellular functions. In this study, we examined the microtubule-binding and assembly-promoting activities of the structural MAPs bound to F-actin. The results showed that actin filament-bound MAP2 and two MAP4 isoforms (LPA5T and LPA4T), but not tau and other two MAP4 isoforms (LPA3T and SPA5T), retained microtubule-binding activity. On the other hand, all microtubule-bound MAPs could not bind to F-actin.

1Pos127 心臓組織片による自律拍動の同期化メカニズム
Synchronization process of cardiac tissue fragments

Tomonori Takahashi[1], Kentaro Ishida[1], Tomoyuki Kaneko[2], Toshiyuki Mitsui[1] ([1]*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*, [2]*Bioscience and applied chemistry, Hosei Univ.*)

The dissected tissue fragments from the heart in sub mm scale exhibit spontaneous contraction. The mechanical-electrical properties, such as interbeat intervals (IBIs), of the fragments differ from their original locations, atrium or ventricle, of the heart. Typically, IBI's of atrium are faster than ventricle's. We have investigated the synchronization processes of the paired fragments, placed next to each other. Initially, each fragment showed different IBI's. However, the pair synchronized their beats after 2:1. Interestingly, the "chaotic states" before the synchronizations were observed. We present the various synchronization processes between various fragments. We also discuss the electrical properties of the paired fragments during their synchronization.

1Pos128 Mycoplasma mobile のストール力
Stall force of Mycoplasma mobile

Masaki Mizutani[1], Yoshiaki Kinosita[2], Takayuki Nishizaka[2], Makoto Miyata[1] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Fac. of Sci., Gakushuin Univ.*)

Mycoplasma mobile, a fish pathogen, forms a membrane protrusion at a pole and glides in its direction. The machinery consisting of 450 units catches sialylated oligosaccharides fixed on solid surfaces. In this study, we focused on "stall force". A polystyrene bead attached to the cell surface was trapped by optical tweezers, slowed down, and finally stalled by a force around 110 pN. A mutant with activated binding was stalled by 90 pN, suggesting that the stall force is determined by the force generation rather than binding. The unit force was detected by the addition of free sialyllactose, distributing around 3.4 ± 0.1 pN. The stall force of a cell may be resulted from the cooperative force generation of 30 units.

1Pos129 定常状態の大腸菌における走化性受容体クラスターの協同性と細胞内シグナル伝達の関係
Relationship between cooperativity in receptor array and intracellular signaling under steady-state of *Escherichia coli*

Hajime Fukuoka[1], Yong-Suk Che[1], Tomoko Horigome[2], Yuichi Inoue[3], Hiroto Takahashi[3], Akihiko Ishijima[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Life Sci, Tohoku Univ.*, [3]*IMRAM, Tohoku Univ.*)

Chemotaxis system is performed by many kinds of proteins. To understand this system as the protein's behavior, we are trying to detect the activity of chemotaxis proteins and the cellular response by the single cell. When receptor (Tsr) mutant, which defects the cooperativity for the attractant response, was expressed with other wild-type receptors, strong dominance negative effect was detected to the switching coordination between flagellar motors in steady-state. This result suggests small number of Tsr mutant collapsed cooperativity of receptor array to affect the CheY-P production. We are constructing fluorescence imaging system to detect the activity of chemotaxis proteins to investigate the cooperativity in receptor array and will show detail in the meeting.

1Pos130 細胞性粘菌における細胞集団の組織的運動の 3 次元解析
3D analysis of collective cell migration in Dictyostelium

Hidenori Hashimura[1], Masato Yasui[2], Kei Inouye[3], Masahiro Ueda[1] ([1]*Department of Biological Sciences, Graduate School of Science, Osaka University*, [2]*RIKIEN QBiC*, [3]*Graduate School of Science, Kyoto University*)

Collective cell movement is involved in various activities such as morphogenesis in multicellular organisms, but the mechanism of the organization of cell movement is still unclear. Dictyostelium cells aggregate by chemotaxis and form multicellular 'slugs' on starvation. The slug migrates straightly and changes migrating direction in response to various stimuli such as light. To reveal how individual cell movement causes the movement of the entire slug, we have developed a new tracking system which enables us to trace almost all cells in the slug three-dimensionally. This method showed the difference in the pattern of cell movement between anterior and posterior cells. Also, we discuss about the change in cell movement when the slugs change the moving direction.

1Pos131 細胞性粘菌の高圧処理からの回復過程で糸状仮足突出が増加する
Protrusion of filopodia increase in the recovery process from the high-pressure treatment of *Dictyostelium* cells

Yuki Gomibuchi[1], Masayoshi Nishiyama[2], Kaoru Katoh[3], Taro Uyeda[3], Takeyuki Wakabayashi[1] ([1]*Teikyo Univ.*, [2]*Kyoto Univ.*, [3]*AIST*)

The effects of high-pressure on *Dictyostelium* cells were examined using a high-pressure microscope. During high-pressure treatment, the shape of the cells became spherical. Contractile vacuoles became less mobile and less obvious. The cells in the recovery phase after the treatment (60 MPa, 2 min) was observed using a usual microscope. Cells tended to show thin filopodia-like protrusions. The cells carrying plasmids to express Tyr143Phe-actin showed more protrusions than those carrying plasmids to express the wild-type actin or the plasmid-free wild-type cells. In filopodia-like protrusions, the fluorescence of GFP-actin was observed. Also, GFP-myosinVII was observed at the tips of the filopodia-like protrusions. Thus, thin protrusions were identified as filopodia.

1Pos132 損傷時の脳がん幹細胞の膜タンパク質の運動解析
Molecular dynamics of brain tumor stem cell induced by specific cell damage

Morito Sakuma, Sayaka Kita, Hideo Higuchi (*Grad. Sch. Sci., Univ. Tokyo*)

Brain tumor stem cells (BTSCs) is recently in the limelight because BTSCs showed distinctive characteristics such as high resistance to therapy, invasion, metastasis, multiline age and self-renewal potential. For removing the brain tumor, the therapy resistance of BTSCs should be evaluated and compared with normal tumor cells. Thus, we selectively damage BTSCs and normal tumor cells by laser activation of near-infrared dyes. We observed the dynamics of membrane proteins and intracellular granules of the damaged cells with a confocal microscope. The motility of membrane proteins and granules in damaged BTSCs was more accelerated than that of normal tumor cells, suggesting that dynamics of proteins would be a key property in therapy resistance of BTSCs.

1Pos133 Local intracellular temperature increase mediates stress granule formation

Beini Shi[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Graduate School of Pharmaceutical Sciences, the University of Tokyo*, [2]*JST-PRESTO*)

Under stress, eukaryotic cells form stress granule (SG) that contains mRNA and translation-related factors. Although SG is recognized as a transient complex responsible for translation reprogramming, the mechanism of assembly is yet to be known. In this study, we focused on the intracellular temperature change induced by various stress during SG formation, and hypothesized that the heat released from mitochondria might play a critical role in triggering SG assembly. Given the results that SG formation could induced by both intracellular thermogenesis and artificial local heating, we confirmed that the intracellular temperature increase is both necessary and sufficient for SG formation, which may be a novel mechanism of this phenomenon.

1Pos134 蛍光イメージングによる単一ミトコンドリアマトリクス内 ATP 濃度測定
Single mitochondrion imaging of ATP concentration changes in the matrix

Saki Yamashita[1], Takahiro Shibata[1], Kotoe Hirusaki[1], Kaoru Katoh[2], Yoshihiro Ohta[1] ([1]*Grad. Sch. Life Sci. & Bio Tech., TUAT*, [2]*AIST*)

Mitochondria are organelles to supply ATP to the cytosol. This process consists of two processes; 1) exchange transport of ADP and ATP and 2) generation of ATP in the matrix. This study was performed to separately observe the two processes in a single mitochondrion. Mitochondria expressing GO-ATeam, a FRET-based ATP probe, were isolated by gentle disruption of cells treated with streptolysin-O and adsorbed onto a glass base dish. Addition of ADP to mitochondria induced the decrease in the FRET efficiency in the matrix. Further addition of malate, glutamate and phosphate increased the FRET efficiency in the matrix. These results suggest that the exchange transport of the nucleotides and the generation of ATP were separately observed in a single mitochondrion.

1Pos135 らせん細菌スピロヘータの調和された回転運動
Coordinated cell rotation in the spirochete Leptospira

Kyosuke Takabe, Md. Shafiqul Islam, Shoichi Toyabe, Seishi Kudo, Shuichi Nakamura (*Grad. Sch. Engineering., Univ. Tohoku*)

Leptospira are spirochetes bacteria comprised of a right-handed helical cell body (protoplasmic cylinder, PC) and two intracellular flagella. Leptospira bend the anterior and posterior ends to spiral-shaped (S) and hook-shaped (H), respectively, for translation. Although rotations of PC and S/H ends due to flagellar rotation are responsible for propulsion, their coordination still remains unclear. Here we simultaneously measured rotation speeds and directions of the three parts in individual swimming cells. Our observations showed the possibilities that the S-end rotation affects the H-end rotation, but frequent independent torque generation of H-end causes transformation from H to S. PC appears to be rotated by counter torque in balance with the S/H ends torque.

1Pos136 Computer simulation of the periodic dynamics of actomyosin foci in C. elegans embryos

Masashi Fujita[1], Shuichi Onami[1,2] ([1]*RIKEN QBiC*, [2]*NBDC, JST*)

Cortical network of F-actin and myosin II often forms focal structures that periodically assemble and disassemble during tissue morphogenesis. The molecular mechanism that produces the periodic dynamics is still unclear. We have experimentally found that a RhoGAP protein RGA-3 has significant effects on the spatiotemporal organization of actomyosin foci in C. elegans. Ectopic localization of RGA-3 results in either oversized or non-pulsatile actomyosin foci. We are now seeking the theoretical framework that explains the actomyosin dynamics of wild-type and mutants in a unified manner. To this end, we modified existing mathematical models of actomyosin by incorporating the effect of RhoGAP. We will discuss some success achieved by our model and its current limitations.

1Pos137 線維芽細胞を介した心筋細胞集団の同期
Synchronization between large clusters of cardiomyocytes through fibroblasts

Shota Miyakoshi[1], Toshiyuki Mitsui[2], Tomoyuki Kaneko[1] ([1]*LaRC, Grad. Sci. Eng., Hosei Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Univ.*)

To analyze the synchronization process of clusters of chicken embryo-derived cardiomyocytes, we used the on-chip multi electrode arrays (MEA) system with agarose microchambers. Two sets of cardiomyocyte clusters were connected with fibroblasts by additional agarose micro fabrication during cell culture. As a result, they were synchronized within the fibroblast distance of 300 μm, and pace making is not depended on the fast beating rate or the stability of fluctuation. It was suggested that another mechanism of synchronization between large size clusters of cardiomyocyte exists different from tissues or single cardiomyocytes.

1Pos138 多細胞の協同運動における力学量の計測と制御
Measurements and control of cellular mechanical factors during collective cell migration

Takeomi Mizutani, Hisashi Haga, Kazushige Kawabata (*Faculty of Advanced Life Sciences, Hokkaido University*)

Multi-cellular migration (collective migration) is an important phenomenon in developmental biology. However, the mechanism is unclear. To understand the mechanism, we focused on cell force. Force map for an epithelial colony showed the region of the intense force did not always correspond to the region of initiation of the collective migration but spatiotemporally varied. Inhibition of cell force within a colony slowed migration speed. Spatial inhibition of cell force at the region of initiation of the collective migration did not hinder cell migration. These results indicate that generation of cell force at the region of the initiation of collective migration is not essential but cell force from the other regions is required for collective cell migration.

1Pos139 海洋性ビブリオ菌のべん毛形成を制御する DnaJ ファミリータンパク質 SflA の構造機能解析

Structural and functional analysis of the DnaJ family protein SflA, that is involved in regulation of flagellation in Vibrio alginolyticus

Satoshi Inaba[1], Takehiko Nishigaki[1], Shoji Nishikawa[2], Mayuko Sakuma[1], Seiji Kojima[1], Katsumi Imada[2], Michio Homma[1] ([1]*Div. Biol. Sci. Grad. Sch. Sci. Nagoya Univ.*, [2]*Grad. Sch. Sci. Osaka Univ.*)

Marine bacterium *Vibrio alginolyticus* usually has a single polar flagellum, and its number and location are regulated by FlhF and FlhG. The Δ*flhFG* mutant has no flagellum, but mutation in *sflA* restores flagellation. SflA is a single transmembrane protein and localizes at the cell pole, but its actual role is unknown. To elucidate the function of SflA, we solved the crystal structure of the N-terminal region of SflA ($SflA_N$) at 2.2 Å resolution. $SflA_N$ forms a dimer in the crystal. The structure of $SflA_N$ is similar to the Tetratricopeptide repeat (TPR) domain, suggesting that this region is involved in protein-protein interaction, although the binding partner of SflA has not been known yet.

1Pos140 微小管を内包した巨大リポソームは架橋・枝分かれ因子無しでも自発的に多角形的な形状になる

Giant liposomes containing microtubules spontaneously develop into polygonal shape in the absence of any crosslinking or branching factor

Masahito Hayashi, Kingo Takiguchi (*Grad. Sch. Sci., Nagoya Univ.*)

We have investigated the various shapes of a giant liposome containing microtubules as a simple mechanical model of a living cell. Although most liposomes smaller than 10 μm in diameter became a symmetrical bipolar shape with a single bundle of microtubules, many liposomes larger than 50 μm became various asymmetrical shapes such as triangles, polygons and spheres with many spikes like a sea urchin. In the polygonal liposome, several bundles of microtubules spanned between the apexes beneath the membrane. This indicates that the mechanical system of cytoskeletal filaments in a membrane container can spontaneously develop into various asymmetrical shapes as well as symmetrical one even without any crosslinking or branching factor of the filaments.

1Pos141 細胞における多種分子活性化の同時計測

Simultaneous detection of the activation of signaling proteins in carcinoma cell lines

Hiraku Miyagi[1], Michio Hiroshima[2], Atsushi Mochizuki[1], Yasushi Sako[1] ([1]*RIKEN*, [2]*RIKEN QBiC*)

The ErbB-RAS-MAPK network is a well-known information transfer system responsible for carcinogenesis. To compare the signaling characteristics of the system between the cell lines established from human carcinoma, we detected activation of multiple species of signaling molecules simultaneously in the same cells stimulated with extracellular ligands for ErbB. We applied immunoblotting analysis in cell populations and found distinctive responses depending on the cell lines and ligand species. We also developed experimental techniques to observe the activation of multiple molecules simultaneously in fixed and living single cells using immunostaining and fluorescent protein tagging, respectively. Differences in the network behavior shown by the methods will be discussed.

1Pos142 心筋細胞に対する力学的刺激の与える影響

Influence of mechanical stimulus on heart cell aggregates

Shin Arai, Ayaha Tsuyuki, Takashi Nakamura, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

The aggregations of cardiac cells spontaneously beat on a petri dish. The numerous studies of their electrical-mechanical interactions with different interbeat intervals (IBIs) have been motivated by potential application in understanding cardiac arrhythmias. To discrete the mechanical interaction, we have applied an artificial mechanical stimulus on single aggregates to excite their beats. The heart cell aggregates were prepared according to traditional protocols. Tungsten probes were used to apply mechanical stimulation cycles on an aggregate. The mechanical stimulation increased the beating activity of aggregates, for example, 66% of the aggregates decrease the IBIs. Furthermore, 34 % of the static aggregates started to beat.

1Pos143 ^{31}P 固体 NMR によるペプチドホルモングルカゴンのアミロイド線維形成過程での生体膜との相互作用変化の解明

Aging in interaction of glucagon with DMPC lipid bilayers in the process of fibril formation as revealed by ^{31}P solid-state NMR

Kazumi Haya, Akie Kikuchi, Izuru Kawamura, Akira Naito (*Grad. Sch. Eng.,Yokohama Natl. Univ.*)

Glucagon is secreted in the pancreatic α-cells and consists of 29-amino acid peptide hormone to control the level of blood sugar. In acidic solution, it is known that glucagon easily forms amyloid fibrils. Temperature variations of ^{31}P solid-state NMR in the glucagon-membrane system were measured. The results indicated that glucagon caused membrane disruption below liquid crystalline-gel phase transition temperature at an early stage in the neutral membrane. This extent of disruption was reduced at the later stage, and we finally observed fibrils with neutral membrane particles by transmission electron microscope (TEM). Time-dependent changes in the glucagon-membrane system indicate that the morphology of glucagon is strongly associated with toxicity of the fibril.

1Pos144 Structure-property of ganglioside GM3/DPPC membranes using coarse-grained molecular dynamics simulation

Kento Inoue[1], Eiji Yamamoto[1], Daisuke Takaiwa[2], Kenji Yasuoka[2], Masuhiro Mikami[2] ([1]*Grad. Sch. Sci. Tech., Keio Univ.*, [2]*Dept. Mech. Eng., Keio Univ.*)

Lipid membranes, including gangliosides, play an important role in various biological processes such as cell adhesion, membrane protein regulation, and virus infection. In this study, we investigate the dependency of structural properties of hydrated ganglioside GM3/dipalmitoylphosphatidycoline (DPPC) membranes on the concentration of GM3 molecules (5, 10, 20 mol %) using coarse-grained molecular dynamics simulations. We observed the formation of raft-like lipid microdomains. We present the effect of these microdomains on the lipid membrane properties, such as order parameter, membrane thickness, area per lipid, and lateral diffusivity of lipids.

1Pos145 逆相法の再検討：ホスファチジルコリン巨大ベシクル形成
A reverse phase method for the formation of giant vesicles (GVs) of phosphatidylcholine

Kanta Tsumoto, Jin Tabata (*Grad. Sch. Eng., Mie Univ.*)

Liposomes are useful for modeling biomembranes, and various preparation methods have been developed based on properties of systems of amphiphiles. Reverse phase evaporation was applied to LUV preparation by Papahadjopoulos [1], and the reverse phase method, which involves w/o/w emulsification, has also been used for GV formation by pioneers [2]. Revisiting its utility, we here examined a simple protocol by which to form egg PC GVs through one-step mixing and centrifugation. We observed many spherical GVs with sucrose (~10%) or salt (~1 M NaCl) in an aqueous phase, though coalescence was enhanced in the latter case.
[1]F. Szoka BBA 601 (1980) 559 [2]S. Kim, G.M. Martin BBA 646 (1981) 1; A.Moscho PNAS 93 (1996) 11443; K. Kato, K. Hirata Solv Extr Res Dev Jpn 3 (1996) 62

1Pos146 化学反応下での脂質二重膜のバディングとコラプス
Budding and collapse of bilayer membrane induced by chemical reaction

Koh Nakagawa, Hiroshi Noguchi (*Institute for Solid State Physics, University of Tokyo*)

In soft matter systems, chemical reaction often induces interesting shape changes (e.g., self-beating gel and self-reproducing vesicle). Such a shape changes are theoretically understood so far. Recently, we studied shape transformations of amphiphilic molecular assemblies induced by chemical reaction as an example of such shape changes using simulation. Coarse-grained molecular simulation technique (Dissipative Particle Dynamics) is used. The simple binding and dissociation reactions are considered as the model of dehydration condensation reaction. Under the chemical reaction, various shape changes are seen in simulation such as collapse and budding of bilayer membrane.

1Pos147 非対称飽和アシル鎖を有するホスファチジルコリンの温度および圧力誘起二重膜相転移
Temperature- and pressure-induced bilayer phase transitions of phosphatidylcholines with asymmetric saturated acyl chains

Masaki Goto, Nobutake Tamai, Hitoshi Matsuki (*Institute of Technology and Science, Tokushima University*)

Most phospholipids in actual biological membranes are asymmetric phospholipids with two inequivalent chains. We have investigated thermotropic and barotropic bilayer properties of asymmetric phosphatidylcholines (PCs) with different saturated acyl chains, the difference of which is two carbon numbers between the sn-1 and sn-2 acyl chains, and found that the mismatch of acyl chain length in terminal methyl ends between both acyl chains produces significant influences on the bilayer properties. In this study, we extend our study to other asymmetric PCs that have different saturated acyl chains by four carbon numbers. We comprehensively discuss the characteristics of asymmetric PC bilayers in comparison with those of symmetric PC and previous asymmetric PC bilayers.

1Pos148 ホスファチジルエタノールアミンを用いて調製したリポソームの特徴
Characteristics of liposomes containing phosphatidylethanolamine

Hayato Akizuki, Tomoyuki Kaneko (*LaRC, Grad. Sci. Eng., Hosei Univ.*)

Liposomes are vesicles of lipid bilayer membrane and used in study as a model of biological membranes. Phosphatidylethanolamine (PE) is a major phospholipid of most prokaryotes. Liposomes were prepared by a lipid film hydration method and varied the ratio of the lipids to clarify whether PE affects the size of liposomes.
As a result, the liposomes containing PE were observed more and smaller than the liposomes not containing PE. Furthermore, the liposomes reproducing the lipid composition of the cell membranes of E. coli were obtained more and smaller than the liposomes reproducing the lipid composition of the cell membranes of human red blood cells. Therefore, it was suggested that PE decreased the size of cells.

1Pos149 分子動力学計算によるセラミド/水界面の水和構造解析
Hydration structure at ceramide/water interface: A molecular dynamics simulation study

Suyong Re, Wataru Nishima, Tahei Tahara, Yuji Sugita (*RIKEN, Wako*)

The water structure at a ceramide/water interface has been studied by using all-atom molecular dynamics simulation in order to understand the stability of ceramide bilayers. The results show that water molecules orient distinctly around each polar region, amide and two hydroxyl groups. Overall, the interfacial water orient as “H-up” in which one of hydrogen atoms of water is head toward the lipid head group. The result is consistent with the recent experimental finding from the heterodyne-detected vibrational sum frequency generation spectroscopy. The presence of distinct local orientation suggests that water molecules contribute to stabilize an inter-lipid network structure of ceramide bilayers.

1Pos150 短鎖リン脂質の吸着膜およびミセルに対するアデノシンリン酸の親和性に及ぼすリン酸基の影響
Effect of phosphate groups on affinities of adenosine phosphates to adsorbed film and micelles of short-chain phospholipid

Ayumi Nishimaru, Miki Tanaka, Michio Yamanaka (*Fac. Sci., Kyushu univ.*)

The surface tensions of aqueous mixtures of diheptanoylphosphatidylcholine (PC) with adenosine phosphates (AP’s), and adenosine, were measured. The phase diagram of adsorption, consisting of the curves of the total molality vs bulk and surface compositions of the mixtures at constant surface tension, and that of micelle formation, consisting of the curves of the critical micelle concentration vs bulk and micellar compositions, were drawn. The shapes of the diagrams indicated the slight incorporation of AP’s and adenosine into the adsorbed film and micelles of PC and the slight difference in the magnitude of the incorporation. These facts suggest that the affinities of AP’s to the adsorbed film and micelles of PC are affected by the phosphate groups of AP’s.

1Pos151 Effect of Osmotic Pressure on Constant Tension-Induced Pore Formation in Lipid Membranes

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Constant tension (σ) in a lipid membrane due to aspiration of a giant unilamellar vesicle (GUV) into a micropipette induces pore formation in the membrane [1]. To elucidate the effective tension due to osmotic pressure (Π), we investigated the effects of Π on the rate constant (k_p) of σ-induced pore formation in a GUV. First, Π was applied to a GUV by its transfer to a hypotonic solution. Then, after the increase of σ to its target, pore formation occurred stochastically, which statistical analysis provided the value of kp [1]. At σ = 0.5 mN/m, k_p values increased with an increase in Π. The effective tensions due to Π obtained by the analysis of the results, were a little smaller than the theoretical values. We discuss its deviation.

[1] Langmuir, 29, 3848, 2013.

1Pos152 抗菌ペプチド・ラクトフェリシンBが誘起する巨大リポソーム中のポア形成
Antimicrobial Peptide Lactoferricin B-Induced Pore Formation in Single Giant Unilamellar Vesicles

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Bovine lactoferricin (LfcinB) has a strong bactericidal activity, but its mechanism is unknown. We found that LfcinB induced rapid influx of membrane-impermeant florescent probe, SYTOX green, from the outside of *E.coli* to its cytoplasm. To elucidate its mechanism we investigated interaction of LfcinB with DOPG/DOPC-membranes using the single giant unillameler vesicle (GUV) method. We observed that a rapid leakage of calcein from a GUV started stochastically, and its statistical analysis provide the rate constant of the LfcinB-induced pore formation. Observation using phase contrast microscopy revealed that LfcinB-induced large pore formation in single GUVs. Based on these data and other experimental results we discuss the mechanism of LfcinB-induced pore formation.

1Pos153 脂質膜表面上での自己組織化された DNA ナノ構造体の構築
Construction of self-assembled DNA nanostructures on lipid membrane surface

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The association between the lipid membranes and the membrane skeleton is important to cell function such as cell signaling. Lipid raft is also important to cell signaling, which is considered to be a form of order-disorder phase separation in model membrane systems. However, the effect of membrane skeleton to phase separation manner is still unrevealed because reconstitution of membrane skeleton in the model system is difficult and ongoing. To elucidate a role of the membrane skeleton in the membrane phase separation, we used DNA nanostructures to imitate biological systems for understanding the living system. Here, we present that a DNA nanostructures as membrane-scaffolding tools is self-assembled on an inner surface of water-in-oil microdroplets.

1Pos154 バクテリアを封入したリポソームにおける大腸菌の動態
Dynamics of Escherichia coli in bacteria-enclosed liposomes

Hazuki Terajima, Ryutaro Isobe, Tomoyuki Kaneko (*LaRC., Dept. Frontier Biosci., Hosei Univ.*)

To give motion ability to the liposome, we tried to be enclosed bacteria into the liposome. At first, we examined the formation ability of liposome in the medium of bacteria. Liposomes above 5μm diameter was able to be made by the lipid composition of phosphatidylcholine (PC), phosphatidylglycerol (PG), and phosphatidylserine (PS). Next, we observed some E. coli was contained inside the liposome. As a result E. coli could not break the membrane of liposomes, and moved usually along the membrane of them. In the future, we will try to examine that E. coli having negative charge attached to cationic lipid (e.g. DOTAP).

1Pos155 リポソーム融合の光マニピュレーション
Photocontrol of liposome fusion

Yui Suzuki[1], Kazuki Shigyou[1], Ken Nagai[1], Anatoly Zinchenko[2], Tsutomu Hamada[1] ([1]*Sch. of Materials Science, JAIST*, [2]*Grad. Sch. of Environmental Studies, Nagoya Univ.*)

Although membrane fusion is the essential event involved to fertilization, intracellular traffic, and viral infection, its biophysical mechanism is poorly understood. In biological systems, the process of membrane fusion is accomplished with the aid of proteins. Recently, in simulation, the change in membrane tension is shown to lead to the fusion of lipid vesicles[1]. Here, we incorporated liposomes with a photo-sensitive surfactant which trigger a change in membrane tension under UV irradiation, and established the photocontrol system of liposome fusion. In this session, we discuss the physical mechanism of the fusion event in terms of the elastic free energy of the membrane. [1] L. Gao, et al., Soft Matter 4, 1208. (2008)

1Pos156 Simple method for lipid bilayer formation with simultaneous incorporation of ion channels using gold electrode (2)

Toru Ide[1,2], Daichi Okuno[2], Takamitsu Kira[1], Minako Hirano[2,3], Hiroaki Yokota[3] ([1]*Grad Schl Natl Sci Tech, Okayama Univ*, [2]*Riken*, [3]*GPI*)

We have developed a technique in which bilayers are made and channels are reconstituted into the membranes by contacting an electrode to a lipid-solution interface. Using this technique, we measured several types of channel which were immobilized on the the electrode. We applied this technique to make a multi-channel measurement system.

The artificial bilayer technique is commonly used to observe properties of ion channels. It allows easy control of the solution and lipid composition. However, while compatible with high-throughput sensor devices for pharmacological screening, the technique is limited to the laboratory due to membrane fragility and a low channel incorporation rate. The simplicity allows our technique to potentially be combined with screening devices.

1Pos157 高速原子間力顕微鏡で見る膜中カリウムチャネル KcsA の動的挙動

Dynamic behavior of the KcsA potassium channel in membrane observed by high-speed atomic force microscopy

Ayumi Sumino[1,2], Takayuki Uchihashi[3], Daisuke Yamamoto[4], Masayuki Iwamoto[2], Takehisa Dewa[5], Shigetoshi Oiki[2] ([1]*JST/PRESTO*, [2]*Facult. Med. Sci., Univ. Fukui*, [3]*Depart. Phys., Kanazawa Univ.*, [4]*Facult. Sci., Univ. Fukuoka*, [5]*Grad. Sch. Eng., Nagoya Inst. Tech.*)

The KcsA potassium channel is a pH-dependent channel, and the activation gate opens at acidic pH. Using atomic force microscopy (AFM), we have captured the conformational change and gating-coupled clustering-dispersion of membrane-embedded KcsA channels. At neutral pH, the closed channels formed nanoclusters. At acidic pH, the nanoclusters disassembled to form individual (or isolated) channels. High-speed AFM captured channel-channel interaction at sub-molecular resolution. At acidic pH, the repulsive motion between the channels was frequently observed. We expect that such dynamic behavior of the KcsA channel in lipid membrane provides insight into mechanism underlying cooperative interplay between ion channels.

1Pos158 ポリセオナミドBチャネルの pH 依存性ゲーティング機構

pH-dependent gating of the polytheonamide B channel

Yuka Matsuki[1], Masayuki Iwamoto[1], Shigeki Matsunaga[2], Shigetoshi Oiki[1] ([1]*Dept. Mol. Physiol. Biophys., Univ. Fukui Fac. Med. Sci., Univ, Fukui, Japan*, [2]*Lab. Aqua. Nat. Products Chem., Grad. Sch. Agri. Life Sci., Univ. Tokyo, Japan*)

We have studied the channel properties of a cytotoxic peptide, polytheonamide B (pTB), from marine sponge by use of the planar lipid bilayer technique. The pTB channel allows permeation of monovalent cations including proton. In this study pH-dependent gating was examined in HCl solutions. Macroscopic current of the pTB channel was recorded upon the voltage steps, and the time-dependent activation and deactivation were observed. At pH 1.5, the channel opens more at positive potentials, while as the pH becomes acidic, the channel opens more at negative potentials. The reversal of the voltage-dependent gating has never been reported for any other channels. We will discuss the molecular mechanism underlying pH-dependency of the gating for the pTB channel.

1Pos159 細菌べん毛 III 型分泌装置のエネルギー変換メカニズム

An energy transduction mechanism of the bacterial flagellar type III secretion apparatus

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The bacterial flagellum is a long filamentous extracellular organelle for motility. Most of its component proteins are transported across the cytoplasmic membrane through the flagellar type III secretion apparatus. To understand the energy transduction mechanism of type III secretion, we have developed an in vitro transport assay system that enables precise control of the measurement conditions. The major energy source of protein secretion is proton motive force (PMF), but the role of ATP hydrolysis energy is still under debate. Here we show that flagellar proteins can be transported by ATP hydrolysis by FliI without PMF. This result suggests that the type III secretion apparatus achieves a robust protein secretion to use both PMF and hydrolysis energy of ATP.

1Pos160 Zero-Mode Waveguides sealed with artificial lipid bilayer for the analysis of membrane proteins

Keisuke Nagao[1], Toshihisa Osaki[2], Ryota Iino[3], Takayuki Uchihashi[4], Hirofumi Shintaku[1], Hidetoshi Kotera[1], Ryuji Yokokawa[1] ([1]*Kyoto University*, [2]*The University of Tokyo*, [3]*Institute for Molecular Science*, [4]*Kanazawa University*)

Membrane proteins play important roles in many physiological functions. Artificial lipid bilayers formed on micro nano structures are promising platforms for studying membrane proteins. However, active transporters which have low transport rates could not be detected by the typical electrical measurement. For the single-molecule fluorescence observation of active transporters, we integrated nanowell arrays called Zero-Mode Waveguides (ZMWs) and lipid bilayers. Artificial lipid bilayers are formed on ZMWs by the vesicle fusion method. The SiO2 deposition on ZMWs was effective for the formation of lipid bilayers. We evaluated the lipid bilayers by fluorescence observations. The lipid bilayers had a good sealing property and showed the potential utility of this platform.

1Pos161 PI3K と Ras から成るフィードバック回路による自己組織的な PIP3 局在形成の制御

Positive feedback loop composed by PI3K and Ras regulates self-organization of PIP3-enriched domain

Seiya Fukushima[1,2], Satomi Matsuoka[2], Masahiro Ueda[1,2] ([1]*Graduate School of Science, Osaka University*, [2]*Riken Quantitative Biology Center*)

Spontaneous cell migration is induced by self-organization of PIP3-enriched domain on cell membrane that acts as a signal for actin polymerization. To reveal the mechanism, we analyzed membrane binding properties of PI3K by multi-color TIRF microscopy in living cells and found that PI3K translocates to the membrane in an activated Ras dependent manner. PI3K started to accumulate on the membrane before PIP3 accumulation, suggesting that Ras activation triggers the domain formation. On the other hand, spontaneous activation of Ras depends on PI3K activity. Therefore, Ras, PI3K and PIP3 possibly compose a positive feedback loop to amplify local PIP3 fluctuations. We will discuss the molecular mechanism based on a mathematical model of single molecule behaviors.

1Pos162 海馬シナプスへの性ホルモンの急性作用：男性ホルモンと女性ホルモンの比較

Acute effect of estrogen and androgen on hippomcapal synapses

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Sex hormones are locally synthesized in hippocampal neurons in addition to gonads. Recently, accumulating evidence provides the acute effect of sex hormones on synaptic plasticity in the hippocampus.
We revealed that kinase networks, downstream molecules of sex hormone receptors in synapse, were involved in acute modulation of synaptic plasticity. These kinase networks are activated within 2 hours, resulting in actin polymerization and spinogenesis in non-genomic manner.
Concerning the signaling cascade of spinogenesis, difference in kinases activated by androgen (DHT) or estrogen (E2) existed. E2 mainly increased small-head spines (0.2-0.4 μm in diameter) whereas DHT mainly increased large-head spines (> 0.5 μm in diameter).

1Pos163 アルギン酸ゲルを用いた神経シートの作製技術の開発と機能評価

Fabrication and functional evaluation of "neuronal sheet" using calcium alginate gel

Hideyuki Terazono, Hyonchol Kim, Fumimasa Nomura, Kenji Yasuda (*Biomed. info., IBB, Tokyo Med. Dent. Univ.*)

We developed a technique to make a neuronal sheet using alginate-coated culture dish. Alginate forms gel state in the presence of calcium ion and change to sol state by removing the ion with a chelating reagent. Using this property, neurons that could be detached from culture dish keeping the neuronal circuit (neuronal sheet). Using the techniques, we demonstrated the community effect between neuronal sheets by partially overlaying neuronal sheets. In the result, while each neuronal sheet had each original firing pattern, firing pattern was synchronized or created a new pattern by overlaying sheet. This technique allows us to demonstrate the plasticity against perturbations from other neuronal networks and to make an artificial neuronal tissue with layer structure.

1Pos164 β3 チューブリン変異による軸索伸長の異常を、キネシンのサプレッサー変異で回復する

Reversal of axon growth defects in CFEOM3 by suppressor mutation in the kinesin-microtubule interface

Itsushi Minoura[1], Hiroko Takazaki[1,2], Rie Ayukawa[1], You Hachikubo[1], Yoshihiko Yamakita[1,3], Seiichi Uchimura[1], Chihiro Yoshida[1], Tomomi Shimogori[1], Tomonobu Hida[1], Hiroyuki Kamiguchi[1], Etsuko Muto[1] ([1]*BSI, RIKEN*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Sch. Med. Nagoya Univ.*)

Mutations in human β3-tubulin (TUBB3) perturb axon guidance, resulting in human nervous system disorder CFEOM3. Here, we investigated microtubule (MT)-based motility in vitro using MTs formed with recombinant TUBB3. We found that disease-associated TUBB3 mutations R262A and R262H impair the motility and ATPase activity of kinesin. Engineering a mutation to L12 of kinesin surprisingly restores a normal level of motility and ATPase activity on the R262A mutant MTs. In parallel with this in vitro observation, in a CFEOM3 disease mouse model expressing the R262A-TUBB3 mutant, overexpressing the suppressor mutant kinesin rescues axon growth. These findings establish the critical role of R262 for mediating interaction with kinesin, which is essential for axon growth.

1Pos165 培養神経回路網における機能的結合のグラフ構造

The graph structure of functional connections in a cultured neuronal network

Nanami Hirata, Hidekatsu Ito, Wataru Minoshima, Suguru Kudoh (*Kwansei Gakuin Univ*)

Recently, a cultured neuronal network is focused on as a model of the fundamental neuronal circuit in a brain. The function of a neuronal circuit is performed depending on the network structure, especially, the network of functional connections. Thus, we established the method to elucidate the graph structure of functional connections in a cultured rat hippocampal neurons. We calculated conditional probability of evoking action potential at a focused neuron on a specific electrode just after the evoking an action potential at a certain neuron, and extracted the graph structure based on the conditional probability. We also confirmed the graph structure was useful criteria for screening of the drug effective against a neuronal circuit.

1Pos166 リアノジン受容体を介した神経細胞の温度感受性カルシウム放出

Thermosensitive Ca2+ burst in rat hippocampal neurons through ryanodine receptors

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We previously reported thermosensitive Ca2+ dynamics in HeLa cells (Tseeb, HFSP J, 2009) and WI-38 cells (Itoh, BIOPHYSICS, 2014), which are attributable to the altered balance between Ca2+ uptake by SERCA and Ca2+ release from IP3 receptors. Here we studied the thermosensitive Ca2+ dynamics in neurons to develop a novel method controlling neural activities with optical microheating. Ca2+ burst was induced by microscopic heat pulses in rat hippocampal neurons. We suggest a plausible mechanism for the Ca2+ dynamics based on SERCA and ryanodine receptors. We also found that the amplitude of Ca2+ burst became larger as the period of culture passed, suggesting the maturation of individual cells and/or the formation of neural networks increases their thermosensitivities.

1Pos167 滑走細菌 *Flavobacterium johnsoniae* のレール状滑走装置

Rail-like structure in gliding machinery of the gliding bacterium *Flavobacterium johnsoniae*

Satoshi Shibata[1], Akihiro Kawamoto[2], Takayuki Katou[2], Keiichi Namba[2], Koji Nakayama[1] ([1]*Graduate Sch. of Biomedical Science, Nagasaki Univ*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

Cells of *F. johnsoniae* move over surfaces, which is called gliding motility. Gliding motility is unrelated to other well-studied bacterial motility systems. In our recent study we proposed a "helical loop track model", where the SprB adhesion protein is propelled along left-handed helical loop on the cell surface. However, little is known about whole shape of gliding machinery.

TEM and Cryo-EM analyses revealed that a rail-like structure, which forms a complex with SprB filaments, was present under the outer membrane. This structure was observed in none of the *gld* mutants that are defective in both gliding motility and the type IX secretion system. These results suggest that the rail-like structure is a part of gliding machinery.

1Pos168 *In-situ* 光照射固体 NMR による D96N-bR 変異体の光中間体と光反応経路の解析

Characterization of photo-intermediates and photo reaction pathway of D96N-bR mutant by *in-situ* photo-irradiation solid-state NMR

Yuto Otani[1], Arisu Shigeta[1], Yoshiteru Makino[1], Ryota Miyasa[1], Izuru Kawamura[1], Takashi Okitsu[2], Akimori Wada[2], Satoru Tuzi[3], Akira Naito[1] ([1]*Grad. Sch. Eng, Yokohama Natl Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Univ. of Hyogo*)

Bacteriorhodopsin (bR) in the purple membrane of *Halobacterium salinarum* exhibits a light-driven proton pump activity. In D96N-bR mutant large dynamic conformational changes in the cytoplasmic region and elongation of half-life of a M-intermediate have been observed during green light irradiation. We stationary observed the photo-intermediates using *in-situ* photo-irradiation solid-state NMR. Using [14, 20-^{13}C]retinal D96N mutant, ^{13}C NMR signals of AT- and CS-state were observed in the dark at -60°C. CS*-, M-, N-, and O-intermediates were detected under irradiation with 520 nm LED light. The photo reaction pathway was estimated as CS→CS*→O, and AT→M→N→O. Thus O-intermediate was observed on light adapted state on transformed from M- and N-intermediates.

1Pos169 局所的な照明により制御される IV 型線毛の収縮
Retraction of Type IV pili controlled by local light gradient

Daisuke Nakane, Takayuki Nishizaka (*Dept. of Phys., Gakushuin Univ.*)

Synechocystis sp. PCC 6803 is a cyanobacterium with the size of 1.5 μm and shows phototaxis as demonstrated in colony spreading on agar. It is hypothesized that the motility is powered by the repetition of the extension and retraction of multiple filaments called type IV pili by energy of ATP hydrolysis. To uncover the mechanism of its signal transduction process of the light-sensing motility apparatus, we here visualized retraction of pili by using fluorescent beads and observed its response to the light. The retraction activity was controlled more effectively by the local light gradient at 300 nm wide rather than lateral light, suggesting that the cell may recognize light direction by the small difference of light intensity around its cell body.

1Pos170 *Harorubrum* 族の新種から見つかったアーキロドプシンの大腸菌での発現
Archaerhodopsin found in *Harorubrum* sp. ejinoor was functionally expressed in *Escherichia coli*

luomeng Chao[1], Xiong Geng[1], Gang Dai[2], Takashi Kikukawa[3], Tatsuo Iwasa[1] ([1]*1Div. Eng. Composite Funct., Muroran Ins. Technol., Japan*, [2]*Coll. Chem. Environ. Sci., Inner Mongolia Normal Univ., China*, [3]*Grad. Sch. Life. Sci., Hokkaido Univ., Japan*)

Haloarchaea *Halorubrum* sp.ejinoor (*He*) was isolated from Ejinoor salt lake in Inner Mongolia of China. Three ORFs of retinal proteins (AR-like, HR-like and SRII-like) were identified in *He* by genome DNA analysis. The real-time RT-PCR study showed that the all genes were expressed. Measurements of light-induced ion transportation of membrane vesicles indicate the existence of a proton pumping and chloride pumping activities. The light-induced absorbance change of *He* membrane showed the presence of the M and O-intermediates. To clarify the physiological functions of the retinal proteins of *He*, we tried to express the genes in *E. coli*. In the present work, the expression and physiological properties of *He*AR are presented.

1Pos171 ニワトリクリプトクロム 4 の光反応の特性解析
Characterization of photoreaction property of chicken cryptochrome4

Hiromasa Mitsui, Keiko Okano, Toshiyuki Okano (*Dept. Eng. and Biosci., Grad. Sch. Adv. Sci. and Eng., Waseda Univ.*)

Cryptochrome (CRY), which function as a photoreceptor in plant and insect, is a candidate photoreceptor or light-dependent magnetoreceptor in vertebrates, though photoreceptive property is still remain elusive. Recently, we determined a putative photocycle of vertebrate CRY, chicken CRY4 (cCRY4). In addition, we calculated absolute absorbance spectra and estimated quantum yields of photoreduction of cCRY4 photointermediates, which enabled us to discuss the potential photoreceptive ability of cCRY4. By comparing a structure and a photocycle of cCRY4 with other CRYs, we assumed that glutamic acid adjacent to a chromophore-binding pocket is important for photoreaction. In this study, we will discuss photoreaction property of cCRY4 in detail.

1Pos172 YFP および mCherry と融合させた光制御型 bZIP タンパク質（Photozipper）の解析
Analyses of a light-regulated bZIP protein, Photozipper, fused with YFP and mCherry

Keigo Furuya, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

We constructed the expression system of a basic leucine zipper (bZIP) protein, Photozipper (PZ), comprised of a bZIP domain and a light-oxygen-voltage-sensing (LOV) domain. In the present study, yellow fluorescent protein or mCherry protein was fused at the N- and C-terminus of PZ, and oligomeric structures of the fusion PZs were compared in the dark and light states. Dynamic light scattering and size exclusion chromatography indicated that blue light (BL) induced the dimerization of each fusion PZ and the increment of its affinity for the target DNA. In addition, fluorescence resonance energy transfer analysis suggested the formation of heterodimers between each fusion PZ. PZs can possibly provide a new approach for controlling bZIP transcription factors.

1Pos173 水溶液中のオキシルシフェリン光ルミネッセンス過程
Analysis of Photoluminescence Pathways of Firefly Oxyluciferin in Aqueous Solution

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To understand the mechanism of firefly bioluminescence, researchers have investigated the spectroscopic properties of emitter, oxyluciferin.However, the emission process of oxyluciferin is still unclear.The aim of this study is to elucidate photoluminescence process of oxyluciferin in aqueous solution at various photoexcitation energies and pH values.The excitation and emission energies of oxyluciferin and its conjugate acids and bases were calculated by the TDDFT. The pathway at pH>8 consists of the vertical excitation from the ground to the first excited states of dianion, decay to its equilibrium structure of the first excited state and emission from this state.

1Pos174 PYP Binding Protein の Rc-PYP との複合体形成における N 末端領域の役割
The role of N-terminal region of PYP binding protein at the formation of complex with Rc-PYP

Tomoyuki Yasumuro, Yoichi Yamazaki, Mikio Kataoka, Hironari Kamikubo (*Grad. Sch. Mat. Sci., NAIST*)

Interaction mechanism of Rhodobacter capsulatus PYP (Rc-PYP) and its binding partner named PYP Binding Protein (PBP) is of interest for the understanding of the signal-transduction process of PYP as well as PAS protein family. However, the mechanism is still unclear, mostly because of lacking of molecular characterization of PBP. The N-terminal 17-residue region of PBP is easily digested by trypsin, but the proteolysis is protected when PBP forms a complex with Rc-PYP. Since this region is expected to be involved in the interaction sites, alanine scanning is applied to this region. Consequently, we found that F16A depresses interaction affinity. The residue should be a key residue in the N-terminal region of PBP for the interaction.

1Pos175 シアノバクテリアに由来する新規クロライドポンプ型ロドプシンは一残基置換によってプロトンポンプへ変換される

A novel chloride–pumping rhodopsin from cyanobacterium converts to proton pump with a single amino acid replacement

Takatoshi Hasemi, Takashi Kikukawa, Masakatsu Kamiya, Tomoyasu Aizawa, Naoki Kamo, Makoto Demura (*Grad. Sch. Life Sci., Hokkaido Univ.*)

Ion-pumping rhodopsins are now classified into outward H^+ and Na^+ pumps and inward Cl^- pump. They have essentially the same structural fold and commonly utilize the photoisomerization of retinal to trigger the respective photoreactions. Despite these similarities, the successful pump-to-pump conversion had been confined to only the H^+ pump bacteriorhodopsin, which is converted to Cl^- pump by a single amino acid mutation. Here we report a novel Cl^- pumping rhodopsin, which is encoded in soil cyanobacterium *Mastigocladopsis repens* and belongs to a cluster far distant from the known Cl^- pumps. By a single amino acid mutation, This rhodopsin was converted outward to H^+ pump. The details will be showed together with other results.

1Pos176 ナトリウムポンプ型ロドプシン KR2 を基にしたカリウムポンプの創成

Engineering K^+ pumping rhodopsin from KR2

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KR2 was identified as a light-driven sodium pumping rhodopsin of marine flavobacteria *Krokinobacter eikastus*. KR2 outwardly pumps Na^+ and Li^+, while it outwardly pumps protons in potassium, rubidium and cesium solution. Molecular mechanism of ion selectivity in KR2 remains unknown. We found a pore-like structure at the cytoplasmic domain in the structure of KR2. Therefore, the effect of N61 and G263, which seem to determine the pore size, was investigated. When N61 and G263 were mutated to bigger amino acids, some mutants pumped not only Na^+ but also K^+. These results suggest that the ion selectivity in KR2 is determined by the volume of positions 61 and 263. Molecular mechanism will be discussed.

1Pos177 ファラオニスハロロドプシンの細胞質側チャネルに位置する重要残基の探索

Functionally important residues in the cytoplasmic half channel of light-driven Cl^- pump *Natronomonas pharaonis* halorhodopsin

Anna Shimosaka, Takashi Kikukawa, Masakatsu Kamiya, Tomoyasu Aizawa, Naoki Kamo, Makoto Demura (*Grad. Sch. Life Sci., Univ. Hokkaido*)

Based on the M intermediate structure in the presence of azide (Nakanishi et al., Biophys. J. 2013), we made 6 alanine mutants in the cytoplasmic (CP) channel and found that L214A, locating on helix F, shows significantly slow photocycle after L2 formation. Replacements to V, F, C and S also slowed down the photocycle, which were then largely accelerated by additional mutations to neutralize the neighboring K215. This lysine residue faces on the outside of CP channel. Thus, its positive charge may contribute to Cl^- transfer by enhancing the dipole moment of helix F pointing toward the CP side. The photolyzed L214 mutants might cause insufficient conformational change, where K215 residue may directly interact with Cl^- and then hinder the translocation inside the channel.

1Pos178 *Tch. tepidum* 由来 LH1-RC の異常吸収挙動と熱耐性の解明に向けて

Toward elucidating the unusual absorption behavior and enhanced thermostability of the LH1-RC complex from *Tch. tepidum*

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The LH1-RC core complex from thermophilic purple bacterium *Tch. tepidum* is characterized by an enhanced thermostability and a Qy transition at 915 nm for the LH1, ~30 nm red-shifted from those of its counterparts. These properties have been demonstrated to be regulated by Ca ions. Replacement of the Ca with Sr and Ba ions resulted in an LH1 Qy transition at 888 nm. The structure of the native LH1-RC has been determined (*Nature* **508**, 228; 2014), in which the Ca-binding sites have been identified. In this work, crystallizations of the Sr- and Ba-substituted LH1-RCs are reported, and spectroscopic characterization of both the solution and crystal samples will be presented.

1Pos179 好熱性紅色光合成細菌由来 LH1-RC 複合体の金属結合サイトおよび金属—タンパク質間相互作用の観測

Monitoring of metal-binding sites and metal-protein interactions in the LH1-RC complex from thermophilic purple photosynthetic bacteria

Yuki Yura[1], Yukihiro Kimura[1], Yusuke Hayashi[1], Li Yong[1], Moe Onoda[1], Seiu Otomo[2], Takashi Ohno[1] ([1]*Grad. Sch. Agri. Sci., Kobe Univ.*, [2]*Fac. Sci., Ibaraki Univ.*)

Metal-binding sites and metal-protein interactions in the LH1-RC complexes from wild-type (B915) and biosynthetically Sr2+-substituted (B888) Tch. tepidum were investigated. Atomic absorption analyses demonstrated that B915 or B888 bind Ca2+ or Sr2+ in the native state. Isothermal titration calorimetry measurements revealed the number of the binding-site and interaction modes between B915 and B888 for the Ca2+-binding to be different. Sr2+/Ca2+ FTIR difference spectra of B915 indicated that the removal of Ca2+ is predominantly responsible for the metal-dependent conformational changes in B915. Based on the present results, the presence of another metal-binding site was proposed in the Tch. tepidum LH1-RC complex.

1Pos180 多孔性シリカ粒子のナノ空間に埋め込まれたヘリオバクテリア反応中心コアタンパクの安定性と分光学的特性

Stability and spectroscopic characterization of heliobacterial reaction center core protein incorporated into nanoporous silica particles

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Heliobacteria are anoxygenic photosynthetic bacteria that have the type 1 reaction center (RC) made up of two identical core subunits. The homodimeric RC core protein (RCcore) from the thermophilc heliobacteium, H. modesitcaldum, was adsorbed under anaerobic conditions into the nanoporous silica particles (SP). The resultant RCcore-SP conjugate exhibited a similar absorption spectrum to that in solution and retained a stable reduction activity of methyl viologen for at least one month. The decay-associated fluorescence analysis at 77K revealed the efficient energy transfer from the antenna pigments to the reaction center P800 without any involvement of free (B)Chls. No critical damages on the RCcore have been caused upon its adsorption to the nanoporous SP.

1Pos181 極低温顕微鏡を用いた緑化途上トウモロコシ生葉の光合成タンパク質前駆体の空間分布の測定

Cryogenic microscope observations of photosynthetic proteins under the assembly process in greening etiolated Zea mays leaves

Tomofumi Chiba, Hiroshi Fukumura, Yutaka Shibata (*Grad. Sch. Sci., Univ. Tohoku*)

To directly observe the spatial distributions of the precursors to photosynthetic pigment-binding proteins in vivo in greening etioplasts, we froze thin sections of greening Zea mays leaves to stop further development of greening and observed them using a novel cryogenic microscope with an NA of 0.9. We performed polarization anisotropy measurements of etioplasts in the earliest stage of greening, and detected no polarization anisotropy. This result suggested absence of the orientational order in this stage of greening despite the crystalline structure of the prolamellar body accumulating the precursor pigments. In 30-min greening leaves, we observed a strong fluorescence peak at 500 nm, which might be assigned to flavin. We will discuss its origin in the poster.

1Pos182 高酸化状態のマンガンクラスターにおける Ca^{2+}の影響

Influence of Ca^{2+} on spin structure of Mn cluster in high oxidation state

Takahiro Sakai[1], Hiroki Nagashima[1], Hitomi Matsushita[2], Ko Furukawa[3], Hiroyuki Mino[1] ([1]*Grad. Sch. Sci., Univ. Nagoya*, [2]*Sch. Sci., Univ. Nagoya*, [3]*Fac., Sci., Univ. Niigata*)

Photosynthetic oxygen evolution is performed at photosystem II (PS II). Mn cluster in PS II catalyzes water oxidation reaction, through five intermediates called S state ($S_{0\text{-}4}$). Ca^{2+} is an atom included in Mn cluster. It has been proposed that Ca^{2+}-depletion affects the electronic structure of Mn cluster in high oxidation state. In order to clarify the effect of Ca^{2+} in high oxidation state, we investigated Ca^{2+}-depleted Mn cluster in high oxidation state using Q-band pulsed EPR measurements. After illuminating for 1 minute at 273 K, EPR signal around g = 3.2 was detected. The result shows that Ca^{2+} directly affects spin structure of Mn cluster in high oxidation state.

1Pos183 光合成水分解反応における S0→S1 遷移の時間分解赤外分光解析

Time-resolved infrared analysis of the S0-to-S1 transition of photosynthetic water oxidation

Tatsuki Shimizu, Ryo Nagao, Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

Photosynthetic water oxidation takes place at the Mn cluster in photosystem II through the so-called S-state cycle (S1->S2->S3->S0->S1). The reaction mechanism of the S0->S1 transition, the final transition of the cycle, has not been well understood, because of contamination of other transitions at the 4th flash. In this study, we analyzed the S0->S1 transition using time-resolved infrared absorption spectroscopy. We applied 3 flashes followed by dark incubation for 30 min to accumulate the S0 and S1 state, and then the time course at 1400 cm-1 was recorded upon one flash illumination. The signal of the pure S0->S1 transition was obtained by removing the S1->S2 contribution, providing the time constant of the proton-coupled electron transfer of the S0->S1 transition.

1Pos184 FRET を用いた放射線誘発 DNA 脱塩基部位の局在性評価

Localization estimation of abasic (AP) sites in DNA induced by ionizing radiation

Ken Akamatsu, Naoya Shikazono (*Japan Atomic Energy Agency*)

Clustered damage site, that is a DNA region with multiple lesions within a few helical turns, is believed to hardly be repaired. However, chemical and spatial details of them are not known. We have developed a methodology for estimating localization of AP sites using fluorescence resonance energy transfer (FRET). We have recently found that experimentally-obtained FRET efficiencies for the heat-treated AP-DNA correspond to theoretical ones calculated on the basis of exponential distribution. We applied the FRET methodology to a plasmid irradiated with radiations such as 60Co-rays, 4He2+, and 12C5+ ion beam. We found that C ion beam tends to produce clustered damage sites compared with the other radiation sources tested.

1Pos185 酸化ストレスはビタミン D によって誘発される骨髄細胞分化を調整する

Oxidative Stress Modulates Vitamin D-induced Myeloid Cell Differentiation

Hiroaki Tanaka[1], Hiroyuki Kato[1], Omi Nawa[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Mater. & Human Env. Sci., Shonan Inst. of Tech.*)

The aim of the present study was to elucidate the function of reactive oxygen species (ROS) in vitamin D3 (Vit D3) -induced antineoplastic effects and immune enhancement using myeloid leukemia PLB-985 cells as a test model. Cells were treated with 200 nM 1α,25-dihydroxyvitamin D3 (an active metabolite of vitamin D3) for 72 h. ESR spin trapping showed that spin-adduct derived from ROS was generated in Vit D3-stimulated PLB-985 cells. Flow cytometry indicated that Mac-1 α was expressed in Vit D3-treated PLB-985 cells but not in gp91phox knockout cells. The present study indicates that ROS enhances Vit D3-induced cell differentiation. These findings suggest that the modulation of oxidative stress potentiates the therapeutic effect of Vit D3.

1Pos186 大腸菌とマイクロデバイスのハイブリッド型人工細胞の効率改善に向けた条件検討

The suitable condition for the hybrid artificial cell system based on *E. coli* and the ALBiC device

Yoshiki Moriizumi[1], Kazuhito V. Tabata[1,3], Rikiya Watanabe[1,3], Hiroyuki Noji[1,2] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ Tokyo*, [2]*CREST, JST*, [3]*PRESTO, JST*)

We are trying to construct ‘a new artificial cell system’ by using the fusion between a protoplast *E. coli* and a micron-scaled reactor ‘ALBiC’, whose top is sealed with the lipid bilayer. We revealed that the inner condition of the chamber could maintain the cell function, such as the protein synthesis. However, the yield of such chambers is still low, which indicates the low possibility of this system. In this study, we are considering the most suitable condition to increase the possibility of this system. We changed the solvent of the lipid solution (hexadecane, squalene and chloroform) and compared the activity in each condition. We would discuss about their correlation and will also report the ongoing project for the development of the artificial cell system.

1Pos187 鋳型複製する高分子系の数理モデルにおける配列情報の選択
Sequence selection in mathematical model of template replicating polymer system

Yoshiya Matsubara, Kunihiko Kaneko (*Department of Basic Science, The University of Tokyo*)

In the problem of the origins of life, how complex RNA sequence have emerged from soup of several types of nucleotides among primitive self-replicating RNA system is a major problem, because only the RNAs which have complex sequence can function as a catalyst. We examined the sequence selected in the template replication system where polymer is synthesized by stepwise polymerization process. As a result, it is revealed that the sequence selected non-trivially varies in dependent with the volume and monomer concentration. In primitive situation there had been places of various space volume and concentration of nucleotides, so the phenomena is considered as one of the causes of increasing the complexity of the RNA sequence.

1Pos188 Visualization of chromatin dynamics and domains in live mammalian cells

Tadasu Nozaki[1,2], Sachiko Tamura[1], Ryosuke Imai[1], Tomomi Tani[3], Masaru Tomita[2], Takeharu Nagai[4], Yasushi Okada[5], Kazuhiro Maeshima[1] ([1]*Natl. Inst. Genet.*, [2]*Inst. Adv. Biosci., Keio Univ.*, [3]*Marine Biological Laboratory*, [4]*ISIR, Osaka Univ.*, [5]*QBiC, RIKEN*)

Long genomic DNA is organized three dimensionally within eukaryotic cell and proteins search their target sites in such crowded nucleus. Here, we report chromatin behavior in living mammalian cells observed by newly developed single nucleosome imaging method. We identified the difference of nucleosome dynamics in two transcriptional chromatin states; nucleosomes around euchromatin-rich regions moved larger than around heterochromatin-rich regions. Furthermore, single nucleosome imaging enabled us to identify numerous compact clusters of nucleosomes in interphase nucleus and mitotic chromosomes. We propose the variation of chromatin dynamics and structure play important roles in search of genomic information by proteins.

1Pos189 フォールド構造に基づく膜タンパク質の分類
Classification of transmembrane proteins based on the fold structure

Tsukasa Ueno[1], Masami Ikeda[1,2], Makiko Suwa[1,2] ([1]*Biol. Sci., Grad. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Chem. Biol. Sci., Sci. Eng., Aoyama Gakuin Univ.*)

To survey structural distribution of proteins, fold recognition method is one of the best ways of structure classification from amino acid sequences. However this method is only established for soluble proteins and new method, available for membrane proteins (MPs), is strongly required. Therefore we propose the proper threshold of structure classification of MP fold structures. For each 37 MP folds (1,528 PDB chains) classified based on SCOPe database, we performed “all-against-all” sequence alignment and structural alignment, respectively. For all (1,166,628) pairs, we observed the relationships between sequence identity (%) and structural similarity (RMSD: Å), and determined suitable thresholds for discriminate the same folds from the different folds.

1Pos190 複合体モデリングにより発見された新規相互作用面と病気関連変異の関係
Relationship between protein-protein interaction interface predicted by complex modeling and disease-related amino acid variants

Toshiyuki Tsuji, Takao Yoda, Tsuyoshi Shirai (*Nagahama Institute of Bio-Technology and Science*)

Many of the biological molecules such as proteins and nucleic acids are assembled into supramoelcules, which are essential to perform complicated functions in the cell. However, It is difficult to determine the structure of protein complex experimentally. We have reported the supramolecule modeling method using IntAct and PDB in the last annual meeting. A total of 3,197 models of human protein complex have been constructed and 25% of inter-subunit interfaces in the models were not experimentally determined, and 41 disease-related amino acid variants associated with the 26 kinds of human disease were mapped on these model-suggested interfaces. For 10 diseases, the relevant 13 variants were mapped to a protein-protein interface as the first time.

1Pos191 ボツリヌス菌の大型プロジェニター毒素複合体のダイナミクス
Dynamics of the Large Progenitor Toxin Complex of Clostridium botulinum

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Botulinum neurotoxins (BoNTs) are one of the potent toxins in nature but the toxicity is immediately eliminated by the harsh environment in the digestive tract because BoNTs are just a protein. However, BoNTs can get to synaptic junctions thanks to support of other proteins, which can form a large progenitor toxin complex. In order to develop a new drug delivery system for oral administration, it might be very helpful that we understand how BoNTs intrude into the bloodstream. Recently, experimental results suggested that, for binding with the intestinal surface, a structural flexibility is important. However, such a flexibility has been only investigated by experimental methods. Therefore, computational analyses are conducted to evaluate how flexible the structure is.

1Pos192 蛋白質複合体における構造変化のデータベース解析
Database analysis of structural changes in protein complexes

Ryotaro Koike, Motonori Ota (*Grad. Sch. of Info. Sci., Nagoya Univ.*)

Many proteins form macromolecular complexes to function. Analyses on the structural complexes are essential to the understanding of protein function. The Protein Data Bank (PDB) provides multiple structures of a protein complex under different conditions, e.g. ligand-bound and ligand-free states. The comparison of structural complexes clarifies the structural change and the molecular mechanism upon ligand binding. We extensively analyzed a huge number of structural complexes in PDB using two programs, SCPC and Motion Tree, which estimate structural similarity and describe structural change between complexes, respectively. The structural changes of various complexes will be characterized in terms of structural symmetry, interfacial properties and so on.

1Pos193 蛋白質の配列進化における最小作用原理とその変異体解析への応用

Application of the principle of least action to protein sequence evolution for quantifying the effect of mutations

Motoi Taniguchi (*Dept. Biol. Sci., Grad. Sch. Sci., Osaka Univ.*)

The multiple sequence alignment (MSA) of a protein family contains rich information about protein evolution. This study aims to develop a method based on the principle of least action to exploit evolutionary information from MSA for functional analysis of proteins. We first define a time-dependent sequence profile which reflects the evolutionary path of a given "wildtype" sequence. Based on this, we define the Lagrangian with residue frequencies at each site as the "coordinates". Solving the equation of motion with a given “mutant” sequence as a boundary condition gives a hypothetical evolutionary path of that sequence. We analyze how the deviation of the path of a mutant sequence from that of a wildtype correlates with experimentally observed effects of the mutation.

1Pos194 遺伝子発現データと染色体構造データに基づく真核生物の染色体上における遺伝子発現相関の解析

Analysis of correlations between gene expressions and chromosome structures of eukaryotes based on publicly available data sets

Rei Tanikado[1], Akinori Awazu[1,2], Hiraku Nishimori[1,2] ([1]*Dept. of Mathematical and Life Sci., Univ. Hiroshima*, [2]*Research center for the Mathematics on chromatin live Dynamics (RcMcD)*)

Several activities of living systems such as the growth and adaptations against the environmental are exhibited through several gene expression processes. Recent experimental studies of several eukaryotes suggested that not only the transcription network structure but also the chromosome physical structures and dynamics play important roles of gene regulations. In this study, we perform the statistical analysis of publicly available data sets of gene expressions and genome structures of S. cerevisiae (budding yeast), S. pombe (fission yeast), and Arabidopsis thaliana. Through such analysis, we reveal the influences of the genome wide physical properties such as nucleosome distributions and 3-dimensional structures on gene expression processes.

1Pos195 Eddy current flow of probability in stochastic gene expression dynamics in eukaryotes

Bhaswati Bhattacharyya, Masaki Sasai (*Sasai group, Department of Computational Science and Engineering, Graduate School of Engineering*)

Quantitative modelling of gene dynamics in eukaryotes remains a challenging problem, unlike simple bacterial cases. Genes in eukaryotes are regulated not only by the protein-factor binding/unbinding but also by epigenetic mechanisms including modification of histones and chromatin structure change. We develop a model to describe such epigenetic processes as a stochastic dynamics in three or more dimensional DNA state space. Gene switching is described as stochastic transitions among basins and saddles in this multi-dimensional DNA space, and the epigenetic dynamics is represented by non-equilibrium curl flux or eddy current of the probability flow in the DNA space. We discuss how this non-equilibrium dynamics regulates fluctuations of gene activity in eukaryotes.

1Pos196 細胞核の変形運動が核内クロマチン配置に及ぼす効果

Nuclear deformation dynamics induced hetero- and eu-chromatin positioning

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The influences of nuclear active deformation dynamics on the intra-nuclear positioning of hetero- and eu-chromatins are investigated. For this purpose, the behaviors of the chains consisting of two types of regions with high and low mobility confined in a pulsating container are considered, where the regions with high and low mobility and the container describe eu- and hetero-chromatin regions and the nucleus respectively. By Brownian dynamics simulations, we found the positioning of low mobility regions exhibit the transition like that from "conventional" to "inverted" hetero-chromatin positioning observed in the experiment of mouse rod cell when the affinity between these regions and the container periphery becomes weak.

1Pos197 適応度と情報に関するゆらぎ定理

Fluctuation Relations for Fitness and Information

Tetsuya Kobayashi, Yuki Sughiyama (*Institute of Industrial Sciene, University of Tokyo*)

Natural selection is a process in which the organisms with higher fitness are amplified within a population.
Since the pioneering work by Fisher, it has been revealed that the natural selection has intrinsic laws that constrain and relate some evolutionary relevant variables such as reproduction rate, variability, and fitness as is the case with heat, entropy, and free energy in the thermodynamics.
By combining the knowledge of evolutionary theory and stochastic thermodynamics, we reveal fluctuation relations that characterize the behavior of fitness in stochastic environment.
We also show that information can be involved in these relations as it does in the Maxwell demon problem in the information thermodynamics.

1Pos198 細胞の自発運動ダイナミクスの種間共通性

Common aspects of spontaneous cell migration dynamics in different species

Hiroaki Takagi (*Dept. Phys., Nara Med. Univ.*)

Cell migration dynamics was studied by statistical analysis of spontaneous migration trajectories, mathematical modeling and simulation. Super diffusion with velocity memory, multiplicative noise, and multiple timescale dynamics were found in Dictyostelium cell. To verify the generality of these dynamics, migration of mouse T cell was analyzed. Several modes in 2D migration were identified, and the effects of inhibitors of specific molecules on cell migration were quantified. Ex vivo 3D migration was also analyzed, and the applicability of the same type of model as Dicty cell was tested in 2D and 3D migration of T cell. Functional significance of fluctuation in cell migration dynamics is to be discussed.

1Pos199 A stochastic simulation study on circadian oscillation and ATPase activity of KaiC hexamer

Sumita Das, Shota Hashimoto, Tomoki P. Terada, Masaki Sasai (*Department of Computational Science and Engineering, Nagoya University*)

KaiC is a hexameric protein, each of its subunit consists of two ATPase domains: CI and CII. It has been reported that the ATPase activity of KaiC is correlated with the oscillation frequency of the KaiABC system. We developed a model of KaiC single hexamer, which describes four phosphorylation states (p-states) of each CII domain, four nucleotide binding states (n-states) of each CI domain and two conformational states (c-states) of each subunit. Stochastic changes in p- and n-states, coupled with the thermal changes in c-states, are simulated by using the Gillespie algorithm. With this model, we examine the underlying mechanism of coupled oscillations of ATP hydrolysis and phosphorylation/ dephosphorylation of single KaiC hexamer.

1Pos200 Dynamical model of chromosome synapsis formation during meiosis in Eukaryotes

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Eukaryotes exhibit “genetic recombination” during meiosis to keep their genetic diversity. Genetic recombination requires juxtaposition and synapsis formation between homologous loci in maternal and paternal chromosomes all along their length. In the recent studies in several organisms, the characteristic dynamical motions of nucleus and chromosomes are observed during meiotic prophase, which might play important roles in the pairing between homologous loci. In this study, we construct a coarse-grained model of chromosomes at meiotic prophase of yeast. By the simulation, we found the difference of the length among non-homologous chromosomes and the dynamical structural transition of the nucleus provide dominant contributions to pairing of homologous chromosomes.

1Pos201 パルス密度制御による小胞型非平衡開放系リアクタ Vesicular nonequilibrium open reactor regulated by pulse-density modulation

Haruka Sugiura[1], Manami Ito[1], Hiroyuki Kitahata[2], Yoshihito Mori[3], **Masahiro Takinoue**[1,4] ([1]*Dept. Comput. Intell. Syst. Sci., Tokyo Tech*, [2]*Dept. Phys., Chiba Univ.*, [3]*Dept. Chem., Ochanomizu Univ.*, [4]*PRESTO, JST*)

Nonequilibrium chemically-open systems are essential for dynamical behaviors of life systems, and the chemically-open systems have recently been developed based on microfluidic devices. Although those devices succeeded in dynamical chemical reactions including chemical oscillations, precise control of chemical dynamics has never been achieved. Here, we report a vesicular nonequilibrium open reactor based on a droplet microfluidics. The reactor enables precise control of chemical dynamics by well-controlled chemical fluxes into/out of reaction systems, which are regulated by pulse-density modulation of vesicular fusion and fission. We believe that this reactor will promote to construct artificial cell-like systems to understand "What is Life?".

1Pos202 キネシン駆動微小管の非平衡パターン形成 Pattern formation of microtubules driven by kinesin

Sakurako Tanida[1], Ken'ya Furuta[2], Kaori Nishikawa[1], Hiroaki Kojima[2], Masaki Sano[1] ([1]*Grad. Sch. Sci., The Univ. Tokyo*, [2]*NICT*)

Animals like fish often organize the groups and they behave like a large creature overall. Such flocking phenomenon occurs in collection of similar interacting units moving with almost the same velocity. It can be also observed in much smaller systems consist of molecules.
Our experimental system called motility assay consists of only a few components found in a cell: microtubule and kinesin. With a large number of microtubules in this system, emergence of collective behavior of self-propelled filaments would be expected. We observed it to investigate the mechanism of collective motion.

1Pos203 高速走査型イオン伝導顕微鏡の開発 Development of high-speed scanning ion conductance microscopy

Shinji Watanabe[1], Toshio Ando[1,2,3,4] ([1]*Bio-AFM, Kanazawa Univ.,*, [2]*Fac. of Math. & Phys., Kanazawa Univ.*, [3]*Grad. Sch. of Nat. Sci. & Tech., Kanazawa Univ.*, [4]*JST-CREST*)

Scanning ion conductance microscopy (SICM) is a powerful tool for visualizing the nanoscale surface structures of biological samples, as its probe does not have to make contact with the samples. However, its data acquisition rate is too low to capture dynamic structural changes of the sample and hence its application has been restricted. Here, we demonstrate a 100-fold improvement in the data acquisition rate. This high rate was accomplished by the development of a fast ion-current detector with a high signal to noise ratio and a high-speed Z scanner. We will describe these developments in detail and discuss how fast a new SICM system with these components can capture an image.

1Pos204 Target imaging droplet sorting system: a shape identification method for recognition and sort target droplet with cell in real time

Mathias Girault[1], Hyonchol Kim[1,2], Kenji Matsuura[1], Masao Odaka[1,2], Hideyuki Terazono[1,2], Akihiro Hattori[1], Kenji Yasuda[1,2] ([1]*KAST*, [2]*TMDU*)

In microfluidic applications, sorting target droplets containing cell or crystal is a key process. Contrasting to the most of sorting methods, we have developed a multiple droplets sorting system based on the real-time image processing. The advantage of this method is the simple set-up of single cell isolation using water droplets in the oil flow. This approach coupled with low cost liquid electrodes and microscopy provides an ideal sorting method for microfluidic applications for crystal or cells with different morphologies. By using the morphological information of targets in the droplets, a mixture of droplets containing single small beads (4.8 or 7.2um) were discriminated and sorted in real time with a high efficiency of validities, 94.9±2% and 98.2±2%, respectively

1Pos205 金ナノ粒子を用いた 1 細胞局所加熱による細胞内温度の研究
Investigation of intracellular temperature using local heating of a single cell with gold nanoparticles

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Temperature is a basic physical quantity. Previous reports show that cells generate heat by chemical stimulation and that there is intracellular temperature gradient. But the mechanism and the significance of intracellular heat generation are unknown. Therefore, we aimed to observe the cell response by intracellular local heating. Here, we investigated the local heating with gold nanoparticles (GNPs), which have been used for the medical research in tumor ablation by heating. We irradiated GNPs which were incorporated into COS7 cells with a laser and observed temperature increase in cells by a fluorescent polymeric thermometer. This result shows that GNPs have a potential for understanding the cell response to the local temperature change of a single cell.

1Pos206 レンズドファイバを用いた内視鏡型蛍光相関分光装置の開発
Development of an endoscopic fluorescence correlation spectroscopy using a lensed fiber

Johtaro Yamamoto, Masataka Kinjo (*Graduate School of Life Science, Hokkaido University*)

Conventional FCS systems are usually composed of a rigid microscope. An endoscopic fluorescence correlation spectroscopy (ES-FCS) have been newly developed using a lensed fiber, optical fibers whose one end face are processed into lens shape, can focus the excitation laser at near the end face in solutions, and the fluorescence signals are corrected by same end. Therefore, it is expected that the ES-FCS achieves the measurements of biomolecular dynamics inside cells of living tissues which have been difficult by conventional FCS due to its large and inflexible instruments. In this study, ES-FCS measurements of fluorescent solutions will be demonstrated. In future, the biomolecular dynamics in animal tissue will be revealed by in situ measurement using the ES-FCS.

1Pos207 アドレナリンによる培養神経細胞軸索輸送活動度の増加がフロー解析法により定量化された
Flow analysis revealed the activity increase of axonal transport of cultured neurons by Adrenaline

Takashi Katakura, Risa Isonaka, Tadashi Kawakami (*Dept. Physiol., Kitasato Univ. Sch. Med.*)

We used KBI Flow Analysis plugin on Image J in our study. Our new developments of the method are that (1) the use of entire area of the video-image with the mask image eliminates inappropriate noise derived from the non-neural area, rather than the use of partial axon area without mask image, (2) rotation of video clips during the preparation process is abandoned to yieled better quality of the video-image.
We have reported the increase in the number of transporting organelles and the velocity both in anterograde and retrograde directions upon the administration of Adrenaline in cultured DRG neurons by our conventional method. Present flow analysis revealed the significant increase in the activity of axonal transport by Adrenaline(0.1-0.5 mM) compared to control.

1Pos208 細胞内小器官選択的な蛍光分子温度計
Organelle-targeting molecular fluorescent thermometers for living cells

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Fluorescent thermometers for living cells currently reported have only one or two of three major properties; 1) sensitivity (relating to the accuracy of measurements), 2) specificity (robustness against the chemical and physical environments) and 3) targeting ability (controlling the location). Here we report novel thermometers that we identified by using a Diversity Oriented Fluorescence Library (DOFL). These small molecules stain cells instantaneously with a simple procedure, specifically target intracellular organelles and report the temperature with high sensitivity and robustness within the physiological condition [Arai, S., *et al.*, *Sci Rep*, **4**, 6701 (2014); Arai, S., *et al.*, *Chem Commun*, **51**, 8044-7 (2015)].

1Pos209 ストレプトリジン O による膜孔形成の高速 AFM 観察
High-speed AFM Observation of Membrane Pore Formation by Streptolysin O

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Streptolysin O (SLO) is one of pathogenic pore-forming proteins and is produced by group A streptococci, whose infection causes scarlet fever, rheumatic fever, and pharyngitis. SLO monomers (60 kD) assemble on membranes to form membrane pores of 20-30 nm in diameter. The structure of SLO has been determined by X-ray crystallography and electron microscopy. However, the dynamic action of single SLO molecules has not been clarified so far, and hence, a poor understanding of the process of pore formation by SLO. In this study, in an attempt to observe the molecular process of membrane pore formation by SLO, we used high-speed AFM (HS-AFM) that enables us to visualize biomolecules in dynamic action, at submolecular spatial resolution and sub-100 ms time resolution.

1Pos210 Development of a novel user-friendly system for image processing of electron micrographs by integrating web browser and PIONE with Eos

Takafumi Tsukamoto, Takuo Yasunaga (*Kyushu Institute of Technology*)

Eos (Extensible Object-oriented System) is one of the applications for image processing of electron microscopy. In usual cases, Eos works with only character user interfaces or integration by some scripting under the operating systems such as OS-X or Linux, not user-friendly. Thus we extended Eos to a web system independent of OS with graphical user interfaces of Web browser. So far, almost all of system implemented properly, produced image analysis, and provided additional actions for improvement of user experiments, such as showing preview image, executing sequential commands, preserving parameters in the execution and restoring them. Furthermore, we have developed the system emplying PIONE, such that our developing platform works under cloud environments.

1Pos211 X 線自由電子レーザーを用いた低温コヒーレント回折イメージングによる酵母細胞核の三次元構造解析

Three-dimensional structure of yeast nucleus visualized by cryogenic coherent diffraction imaging using X-ray free-electron laser

Yuki Sekiguchi[1,2], Amane Kobayashi[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Yuichi Ichikawa[3], Hitoshi Kurumizaka[3], Mitsuhiro Shimizu[4], Masaki Yamamoto[2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*, [3]*Grad. Adv. Sci. Eng., Waseda Univ.*, [4]*Grad. Sci. Eng., Meisei Univ.*)

Coherent X-ray Diffraction Imaging (CXDI) visualizes electron density distribution inside non-crystalline biological particles with size of micrometer to sub-micrometer without sectioning or staining. In CXDI experiments, particles are irradiated by coherent X-rays and electron density maps projected along the direction of incident X-rays are retrieved from diffraction patterns. 3D structures of particles can be reconstructed from a large number of projected maps. Organelles like nuclei are quite large so that their whole structures have not been revealed well beyond resolutions of optical and fluorescence microscopy. Here we report a reconstructed 3D structure of yeast nucleus at a period of cell cycle by performing CXDI experiments using X-ray free-electron laser.

1Pos212 幅広い応用が可能な超高輝度マルチカラー発光タンパク質

Color pallet of super-duper luminescent proteins capable of wide range application

Kazushi Suzuki[1], Yoshiyuki Arai[1,2], Masahiro Nakano[1,2], Takeharu Nagai[1,2] ([1]*Grad. Sch. Engin., Osaka Univ.*, [2]*ISIR, Osaka Univ.*)

Luminescence imaging is promising bioimaging modality because it circumvents potential problems in fluorescence imaging, such as phototoxicity, and photobleaching. However, dim signal of the luminescence hampers universal applications. To overcome this problem, we have already developed a brighter luminescent protein, Nano-lantern (Saito K. et al., 2012) and its color variants (Takai A. et al., 2015). Here, we report an enhanced version of the Nano-lantern (eNano-lantern), its color variants and Ca2+ indicator, which emit 10 times brighter signal than that of Nano-lantern. We will demonstrate world-first single molecule luminescence imaging as well as multicolor observation of protein dynamics, and long-term ultra-fast visualization of Ca2+ dynamics in cardiomyocyte.

1Pos213 Yeast two hybrid および定量イメージング解析による PDLIM2 相互作用タンパク質の同定

Identification of PDLIM2 interacting protein using yeast two-hybrid system and quantitative imaging analysis

Chanyoung Shin[1,2], Yuma Ito[1], Kumiko Sakata-Sogawa[1], Takashi Tanaka[2], Makio Tokunaga[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*IMS-RCAI, RIKEN*)

PDLIM2 has an important role for inactivation of NF-κB-mediated signaling. However, it remains unknown how PDLIM2 can be controlled. To clarify this mechanism, we sought to identify the molecules that interact with and functionally affect the activity of PDLIM2 protein using a yeast two-hybrid screen. Here we found that six proteins, AP2M1, BSG2, CTSB, DPP9, MKRN2, TBC1D14, could associate with PDLIM2. We further demonstrated that all of these proteins inhibited p65-mediated gene activation in luciferase assay. These results suggest that the PDLIM2-interacting proteins that we identified are novel negative regulators in NF-κB signaling. To elucidate the function of these proteins upon LPS stimulation, we visualize and analyze the dynamics by quantitative imaging.

1Pos214 Measurement of the redox potential in Chlamydomonas flagella using a redox-sensitive fluorescent protein, Oba-Q

Yuta Nishimaki, Kazunori Sugiura, Toru Hisabori, Ken-ichi Wakabayashi (*Chemical Resources Laboratory, Tokyo Institute of Technology*)

The unicellular green alga Chlamydomonas swims with two flagella, in which dyneins generate force for beating. Several lines of experiments suggest that Chlamydomonas flagellar motility is regulated by redox poise; however, whether or not intraflagellar redox state changes has not been quantitatively tested. In this study, we incorporated Oba-Q, a newly developed redox-sensitive fluorescence protein, with Chlamydomonas flagellar axonemes as a fusion protein with a dynein subunit. From a calibration graph of fluorescence vs redox potential established in vitro, we could estimate the intraflagellar redox potential of Chlamydomonas as -275.6 ± 5.5 mV. Currently we are trying to see if it changes under different culture conditions and the results will be discussed.

1Pos215 クロマチン動態制御におけるアクチン関連タンパク質 Arp4 の核内ダイナミクス

Dynamics of actin-related protein 4 in living cell nucleus for dynamic chromatin regulation

Yuma Ito[1], Hiroshi Kimura[1], Masahiko Harata[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

Actin-related protein 4 (Arp4), a key component of chromatin remodeling complexes and histone acetyltransferase complexes, plays an important role in transcriptional regulation. Although previous studies have shown that Arp4 was bound to core histones, its dynamic mechanism remains elusive. To quantify Arp4 dynamics in the nucleus, here we applied an imaging approach in living HeLa cells. Single-molecule imaging and FRAP analysis revealed transient binding of Arp4 to nuclear structures. ATP-binding site mutants and ATP depletion showed increases in Arp4 dynamics. These results suggest that the dynamic interaction requires ATP binding. We will discuss a quantitative model of nuclear Arp4 dynamics related to chromatin remodeling.

1Pos216 蛍光褪色回復法による INO80 クロマチンリモデリング複合体の核内動態解析

Intranuclear dynamics of INO80 chromatin remodeling complex by fluorescent recovery after photobleaching

Yuma Ito[1], **Tsubasa Isogaki**[1], Hiroshi Kimura[1], Masahiko Harata[2], Kumiko Sakata-Sogawa[1], Makio Tokunaga[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

The INO80 chromatin remodeling complex has an important role in transcription, DNA replication and repair by regulating chromatin structure. INO80 contains actin-related proteins Arp4, Arp5 and Arp8. However, the role of Arps in chromatin regulation mechanism is not fully understood. To quantitate the dynamics of Arps, here we performed fluorescence recovery after photobleaching (FRAP) of Ino80, a subunit of INO80 complex, and Arps in the cell nucleus. FRAP curves of Arp4, Arp5 and Arp8 were different from Ino80, suggesting the dynamic formation of the INO80 complex. Quantitative analysis of FRAP curves indicates that Arps and Ino80 interact with the nuclear structure in different manner. We will discuss about the dynamic mechanism of Arps in chromatin remodeling.

1Pos217 高速原子間力顕微鏡により観察された AAA シャペロン p97 の主要 ATPase リングの構造変化

Conformational changes of the major ATPase domain D2 of the AAA chaperone p97 observed by high-speed atomic force microscopy

Daisuke Yamamoto[1,3], Kentaro Noi[2,3], Ken-ichi Arita-Morioka[2,3], Teru Ogura[2,3] ([1]*Fac. Sci., Fukuoka Univ.*, [2]*IMEG, Kumamoto Univ.*, [3]*CREST, JST*)

p97 is an AAA chaperone, which plays crucial roles in a variety of cellular processes. p97 comprises three domains, N-terminal domain and two ATPase domains (D1 and D2), and forms a homo-hexameric ring. Here, we applied high-speed atomic force microscopy (HS-AFM) to directly observe the conformational changes of the major ATPase domain D2 during the cycle of ATP hydrolysis. In the absence of ATP, each subunit and a central pore of the D2 ring were clearly observed. By contrast, in the presence of ATP, the central pore was less visible because of the conformational change of the D2 domain. This conformational change was accompanied by the rotation of the D2 ring relative to the N-D1 ring, which is consistent with our previous HS-AFM observation of the N-D1 ring.

1Pos218 軟 x 線顕微鏡による糸状シアノバクテリアにおける窒素固定の直接観察

Direct observation of nitrogen fixation in filamentous cyanobacteria by using soft X-ray microscopy

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The cyanobacterium Anabaena sp. PCC 7120 is an oxygenic photosynthetic prokaryote that performs nitrogen fixation. It forms a filament consisting of two different types of cells, so-called “vegetative cell” and “heterocyst”, at a ratio of 10:1. Differentiation of heterocyst is believed to be triggered by the carbon-nitrogen ratio (C/N ratio) in the cell; however, the difference of the C/N ratios of individual cells have never been examined directly. In this study we observed the heterocyst and vegetative cells of Anabaena sp. PCC 7120 by using soft X-ray microscopy and directly map nitrogen atoms in each cell by observing at the wavelength shorter and longer than the nitrogen K-absorption edge.

1Pos219 遠心操作を伴わない細胞を回収するためのマイクロデバイス

Microfluidic Device for Recovering Cells without Centrifugation

Koji Matsuura, Saori Nishina, Keiji Naruse (*Cardiovascular Physiology, Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama University*)

To reduce Intracytoplasmic sperm injection treatment time of patient’s sperms, we developed a microfluidic device to increase sperm concentration. The microfluidic chamber was manufactured by a conventional mechanical microdrilling process and was subsequently wrapped with polyvinylidene chloride using a laminating machine. We achieved a 10-13-fold enrichment ratio in nonmotile human sperms (10k cells/mL) when a chamber height of 1.4 mm was used. Similar results of enrichment were obtained using round cells and microparticles with 0.01 mm of diameter. We are currently optimizing microfluidic channel structure to improve the recovered concentration of these cells in higher cell concentration.

1Pos220 芽胞菌に対するマイクロ波照射効果

Microwave Irradiation Effect to Spore-Forming Bacteria

Ryota Nakama[1], Arata Shiraishi[1], Wataru Nagayoshi[1], Sakura Yoshimoto[2], Shokichi Ohuchi[1,2] ([1]*Dept. Lifesci. & Syst, Eng., Kyushu Inst. Tech.*, [2]*Dept. Biosci. & Bioinform., Kyushu Inst. Tech.*)

Bacillus subtilis is used for the substance production from its usefulness. On the other hand, *Bacillus subtilis* forms easily spore state in the unsuitable environment for growth. Spores show high resistance to heat and chemicals, sterilization is very difficult. We have revealed the number of bacteria increased or decreased by the amount of the output of the microwave power. In this study, we developed sterilization technique of spore-forming bacteria under microwave irradiation. We discussed the effect of microwave to sterilization technique.

1Pos221 ビオチン化したタンパク質ラングミュア膜上に作成したストレプトアビジン層

Streptavidin layer formed on the biotinylated Langmuir film of protein

Taiji Furuno (*Dept. Phys., Keio Univ. Sch. Med.*)

A layer of streptavidin was formed via an easy method on the surface of silicon wafer. A surface film of carbonic anhydrase (CA) denatured at the air/water interface was transferred onto the hydrophobic surface of silicon wafer by means of Langmuir-Schaefer technique. The transferred CA film was biotinylated and subsequently allowed to bind streptavidin to form a second layer. Biotinylated proteins, ferritin, catalase, alcohol dehydrogenase and carbonic anhydrase, were incubated with this SAv layer and examined by atomic forces microscopy. Adsorption of these biotinylated proteins at high densities was confirmed, while a subtle or negligible amount of nonspecific binding of intact proteins was observed.

1Pos222 光分解性ゲルによる接着細胞の力学的操作法の検討

Application of photodegradable hydrogel to mechanical manipulation of adherent cells

Kentaro Iketaki[1], Humiki Yanagawa[2], Ryuzo Kawamura[1], Seiichiro Nakabayashi[1], Toshiyuki Takagi[2], Shinji Sugiura[2], Toshiyuki Kanamori[2], Hiroshi Yoshikawa[1] ([1]*Dept. Chem., Saitama Univ.*, [2]*BRD., AIST.*)

Photodegradable hydrogel has been attracting much attention as a powerful experimental platform that can provide dynamic cellular environments. Recently, we proposed a new class of photocleavable crosslinkers that can form photodegradable hydrogel with various biocompatible polymers (e.g., gelatin and matrigel) [1]. So far, the photodegradable hydrogel was applied to the fabrication of 3D microscaffolds and cell culture substrate with tunable stiffness. In this work, we propose a new application of the photodegradable hydrogel to the mechanical manipulation of adherent cells, which can also be used to produce spheroid-like cell aggregates.

[1]F. Yanagawa et al., Adv. Healthcare Mater. 4 (2015) 246-254.

1Pos223 電極埋め込み型ナノポアの AC ゲート電位による DNA の挙動制御

Controlling DNA motions with an AC gate voltage applied gate embedded in nanopore

Yuta Kato, Naoto Sakashita, Yoshitaka Tanida, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

Sensing devices using solid-state nanopores have been developed toward single DNA sequencing as an ultimate goal. One of the main issues concerning the nanopore devices is that the DNA molecules through a nanopore, are too fast. Recent trends of reducing the DNA molecule's speed are implementing AFM or optical tweezers. However, these instrumentations require technical expertise and high installation costs. Here, we report the fabrication of a nanopore with embedded gate electrode, and demonstrate the slowed DNA oscillations via the pore by applying non-sinusoidal AC voltage such as square and sawtooth waves with their frequencies from 0.5 to 10 Hz on the gate. Finally, we present the ratchet-like DNA motions induced by the selected sawtooth waves.

1Pos224 ハニカム構造を有するマイクロゲルネットワークの構築

Construction of microgel network with honeycomb structure

Satoshi Umeyama, Masayuki Hayakawa, Masamune Morita, Masahiro Takinoue (*Interdisciplinary Graduate school of Science and Engineering*)

Artificial-cell networks using aqueous droplets have recently been constructed as a model to understand cooperative behaviors of population of living cells like tissues. However, the aqueous droplet networks have a problem that their structures break by droplet fusion. Here, we constructed an artificial-cell network using a stable honeycomb structure microgel. We changed the size and the packing degree of microgels; in addition, we observed the diffusion in microgel network using fluorescence recovery after photobleaching method (FRAP). We believe that our study will contribute to not only understanding of the system of living cells but also development of functional materials and microdevices such as neural-network-like electrical circuit in a bottom-up manner.

1Pos225 Directed evolution system to generate peptide agonists for G protein-coupled receptors using in vitro translation in water-in-oil droplets

Takashi Sakurai[1], Ryo Iizuka[1], Yasuyuki Nakamura[2], Jun Ishii[3], Rui Sekine[4], Yoon Dong H.[4], Tetsushi Sekiguchi[5], Akihiko Kondo[2], Shuichi Shoji[4], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Eng., Kobe Univ.*, [3]*Org. of Adv. Sci. and Technol., Kobe Univ.*, [4]*Grad. Sch. of Adv. Sci. and Eng., Waseda Univ.*, [5]*Inst. for Nanosci. and Nanotechnol., Waseda Univ.*)

G protein-coupled receptors (GPCRs) are particularly important drug targets in the pharmaceutical industry. Thus, intense efforts have been made to screen new GPCR ligands for potential drug candidates. However, for many GPCRs, such attempts have failed to yield desired results. To overcome the problem, we designed a directed evolution system to generate peptide agonists for GPCRs using water-in-oil (W/O) droplets. To achieve the system, we established basic techniques such as generation of size-controlled W/O droplets using microfluidics, peptide synthesis and agonistic evaluation in the same droplet, and microfluidic fluorescence-activated sorting of W/O droplets. The combination of these techniques will generate novel peptide agonists for many GPCRs.

1Pos226 分子モータと DNA 回路を内包した巨大リポソームにより構築される分子ロボット

Molecular robots constructed by entrapping molecular-motors and DNA circuits into giant liposomes

Yusuke Sato[1], Yuichi Hiratsuka[2], Ibuki Kawamata[1], Satoshi Murata[1], Shin-ichiro Nomura[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Sch. Matl. Sci., JAIST*)

Molecular robotics is a new research field to develop an artificial molecular system. Recently, single-molecular robots with random motion in water solution have been reported. This study aims to create molecular robots with autonomous and controllable mobility. As a design of such molecular robots, we propose an amoeba-like structure. The robot consists of molecular-motors (kinesin/microtubules) with DNA circuits entrapped by giant liposome. It is known that the motors can deform lipid membrane. We found a reasonable condition for entrapping molecular-motors into giant liposomes at an efficiency of more than 50 %. We will discuss about controlling deformation of liposomes by the combination of molecular-motors and DNA circuits.

1Pos227 光応答基板：細胞移動研究のためのマテリアルバイオロジー的研究ツール

Photoactivatable substrates: Material biological tools for cell migration research

Jun Nakanishi[1], Kazuo Yamaguchi[2] ([1]*NIMS*, [2]*Kanagawa University*)

Cell migration is a fundamental cellular activity involved in various biological processes. Understanding its regulatory mechanisms is important not only for basic biological interests, but also for tissue engineering and drug discovery. Our group addresses to cell migration phenomenon by chemically and physically controlling cellular nano- and microenvironments by using originally-developed photoactivatable substrates. The common feature of these substrates is that the surface changes from a state that preventing cell adhesion to that promoting cell adhesion in response to photoirradiation. In this poster, I will introduce some of our recent results on collective migration behavior of the cells under such defined microenvironments.

1Pos228 Novel genetically encoded antibody-based biosensors allowing fluorescence ratio detection of antigens

Kim Phuong Huynh Nhat, Takayoshi Watanabe, Takahiro Hohsaka (*JAIST*)

N-terminus fluorescently labeled single-chain antibodies (scFv) that show antigen-dependent fluorescence change have been developed. However, the N-terminal labeling requires incorporation of fluorophore-labeled non-natural amino acid to scFv in a cell-free translation system. Here, we utilized SNAP-tag protein and its fluorescent ligands for N-terminal fluorescent labeling of scFvs. The SNAP-scFv fusions showed antigen-dependent fluorescence enhancement. In addition, fusions of fluorescent protein (FP) to SNAP-scFvs showed FRET between FP-fluorophore and antigen-dependent change of fluorescence ratio of fluorophore and FP. Our strategy has a wide range of applications since this biosensor is easy to design for various antigens and can be expressed in living cells.

1Pos229 ベイズ推定を用いた CTF 補正の自動化

Automation of CTF correction using the Bayesian estimation

Koji Hisanaga, Takuo Yasunaga (*Kyushu Institute of Technology Graduate School of Computer Science and Systems Engineering Yasunaga Lab*)

Cryo-electron microscopy has been used for observation at the molecular level of proteins in a cell or in vitro. The resolution of three-dimensional maps reconstructed from the obtained two-dimensional projection images has reached a near atomic resolution. However, it is necessary to take defocused projection images for the purpose of improvement of contrast but result in blurring of the image, which is represented as CTF (contrast transfer function) in the Fourier space. It takes much time and effort to determine the defocus values from image and correct blurring of the CTF.
In this research, we will estimate defocus values by using Bayesian estimation, and evaluate automate correction of the CTF.

1Pos230 Model studies of bacterial flagellar motor switching in response to CheY-P regulation and motor structural alterations

Qi Ma[1], Matthew A. B. Baker[2], Fan Bai[1] ([1]*Biodynamic Optical Imaging Center, Peking University, Beijing, China*, [2]*Victor Chang Cardiac Research Institute, Sydney, NSW, Australia*)

The Bacterial Flagellar Motor (BFM) rotates the helical filaments and propels the bacteria swimming towards favorable conditions. Previously we built a stochastic model featuring the "conformational spread" to explain the dynamic behavior of BFM switching. Here we further analyzed this model and showed that 1) the model predicted a strong correlation between motor rotational direction and the number of CheY-P bound to the switch complex; 2) the model also predicted that stoichiometry of FliM underwent dynamic exchange in order to maintain ultrasensitivity in motor switching response; 3) the model simulated the recent experiments expressing CW-locked mutant FliG into the switch complex and reproduced the enhanced switching frequency of the motor.

2Pos001 CryoEM 3D image reconstruction of the flagellar LP ring complex

Meltem Tatli[1], Tomoko Miyata[1,2], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Riken QBiC*)

Many bacteria swim using flagella, each of which is rotated by a rotary motor in the basal body (BB). The LP ring is a molecular bushing, embedded in the outer membrane and peptidoglycan layer for the rod to span these two layers to function as a drive shaft, rapidly rotating without much friction to transmit motor torque to the filament working as a helical propeller. I improved its isolation conditions to efficiently prepare a large amount for cryoEM structural study, but the LP rings tended to form aggregations. So I tried different chemical reagents and detergents to make it monodispersed. I have been collecting cryoEM images for 3D image reconstruction by single particle image analysis to understand the mechanism of the LP ring working as a molecular bushing.

2Pos002 毒性型アミロイドテープ： GM1 クラスター上で形成される新規 Aβ 逆平行／平行混合 β シート構造
Toxic Amyloid Tape: Novel Mixed Antiparallel/Parallel β-Sheet Structure Formed by Aβ on GM1 Clusters

Yuki Okada[1], Keisuke Ikeda[5], Yoshiaki Yano[1], Masaru Hoshino[1], Yoshio Hayashi[4], Yoshiaki Kiso[3], Hikari Itoh-Watanabe[2], Akira Naito[2], **Katsumi Matsuzaki**[1] ([1]*Grad. Sch. Pharm. Sci. Kyoto Univ.*, [2]*Fac. Eng. Yokohama Nation. Univ.*, [3]*Nagahama Inst. Bio-Sci. Tech.*, [4]*Tokyo Univ. Pharm. Life Sci.*, [5]*Grad. Sch. Med. Pharm. Sci. Univ. Toyama*)

The aggregation of Aβ is central in the pathogenesis of Alzheimer's disease. We found that amyloid fibrils formed on GM1 clusters were more toxic than those formed in solution. Less toxic fibrils formed in solution are considered to contain in-resister parallel β-sheets, whereas the structure of the toxic fibrils is unknown. In this study, we investigated the structure of the toxic fibrils in detail. Chemical cross-linking experiments suggested that the fibrils contained both parallel and antiparallel β-sheet structures at an approximate ratio of 2:1. Solid-state NMR and isotope-edited FTIR experiments also supported this conclusion. AFM revealed that the toxic fibrils had a flat, tape-like morphology. Thus, the toxic fibrils possess a novel unique structure.

2Pos003 Hsp90 と ADP の会合における水分子の役割
Role of water molecules for association of Hsp90 and ADP

Kazutomo Kawaguchi, Hiroaki Saito, Hidemi Nagao (*Inst. Sci. Eng., Kanazawa Univ.*)

Heat Shock Protein 90 (Hsp90) is one of a group of molecular chaperones required for protein folding. The functional cycle of Hsp90 is driven by ATP association, hydrolysis of ATP to ADP and ADP dissociation. In our previous study, we calculated the free energy profile for ADP dissociation and found effective attraction by using molecular dynamics simulation [K. Kawaguchi, et al., Chem. Phys. Lett., 2013]. In this study, to investigate the solvent effect for the stabilization of Hsp90-ADP association, the calculated association free energy is divided into the contributions of solvent water and other molecules. It is clearly shown that van der Waals interaction from water molecules strongly stabilizes the Hsp90-ADP association.

2Pos004 赤外分光法によるインスリンアミロイドの構造規則性の解明
Revealing the structural rules of insulin amyloid by infrared spectroscopy

Hisayuki Morii[1], Takashi Shimizu[1], Masayuki Nara[2] ([1]*AIST*, [2]*Tokyo Med. Dent. Univ.*)

Insulin is a disulfide-linked peptide consisting of two peptidyl chains. Previous analyses by 2-dimensional screening suggested that the region of (13-27) in its B-chain was essential for amyloid formation. Thus, a series of site-specific ^{13}C-labeled peptides of this region were synthesized and the resulting amyloid fibrils were analyzed by IR spectroscopy. On the basis of isotope-induced IR band shifts, the secondary structures of respective residues were identified. The molecular structure in amyloids is most likely to be constructed of two β-strands, which are formed only in the amyloids, and a loop connecting them. In addition, the relative alignment of two interacting β-structure regions will be discussed based on the IR analyses of ^{13}C/^{15}N-dually-labeled peptides.

2Pos005 タンパク質の形状に内在する機構的性質に関する解析
Analysis of Mechanism Features Present in Protein Shapes

Keisuke Arikawa (*Fcl. Eng. , Kanagawa Inst. of Tech.*)

Many proteins change their conformations like a mechanism. It can be understood that the mechanism features are present in the shapes of proteins. We developed a method for analyzing mechanism features using an elastic network model (ENM). As the key procedure in the analysis, we sequentially pinched the localized region in ENM and evaluated the deformation of the entire region. The pinched regions that caused larger deformation of the entire region corresponded to the joints of mechanisms. In addition, by applying our abovementioned analysis for both the states before and after interacting with other molecules and comparing the results, we can obtain insights on the structures that attribute to the protein function.

2Pos006 抗体 CDR-H3 ループ領域の立体構造予測
Accurate ensemble modeling of CDR-H3 loop in antibody

Hiroshi Nishigami[1,2], Gert-Jan Bekker[1], Narutoshi Kamiya[1], Junichi Higo[1], Haruki Nakamura[1] ([1]*Institute for Protein Research, Osaka University*, [2]*Department of Biology, Graduate School of Science, Osaka University*)

The antigen-binding site in antibody, CDR, is hypervariable in amino acids sequence. In particular, the third CDR loop of the heavy chain, CDR-H3, is so variable in sequence, length and conformation, that fragment structures in PDB are not enough for its precise structure prediction, although several sequence-structure relationships have so far been proposed. In addition, the flexible nature of the CDR-H3 loops makes it difficult to build a single template-based 3D-model. We executed conformational sampling of a CDR-H3 loop with surrounding residues in explicit solvent by multicanonical molecular dynamics method to find stable structures as an ensemble. The accuracy is evaluated by comparing the structures in the canonical ensemble at 300 K with the crystal structure.

2Pos007 単粒子像解析のための分子同定用 GFP ラベル
GFP protein labeling for single particle image analysis

Takayuki Kato[1], Naoya Terahara[1], Tomoko Miyata[2], Keiichi Namba[1,2] ([1]*Osaka Univ, Front. Biosci.*, [2]*Riken, Qbic*)

The single particle image analysis by cryoEM is very powerful tool for the structural analysis. Recently, some structures have been analyzed beyond 3 angstrom resolution. But in the case of a flexible or a heterogeneous molecule, the resolution is lower than ~15 angstrom. Such a intermediate resolution, the orientation of the molecule or boundary of the each molecule in the molecular complex cannot be even decided. On the other hands, GFP is widely used for various biological phenomena analysis under the optic microscopy. Florescence from GFP cannot be detected by electron microscopy, but extra mass can be visualize. I will discuss about possibility of the GFP labeling to decide the molecular orientation and position for single particle analysis.

2Pos008 広範囲に基質特異性を示す L-アミノ酸酸化酵素の構造
Structure of L-amino acid oxidase with broad substrate specificity

Nanako Ito[1], Tatsuya Kawaguchi[1], Kaho Murakami[2], Takashi Tamura[2], Miwa Yamada[3], Kimiyasu Isobe[3], Kenji Inagaki[2], Katsumi Imada[1] ([1]*Dept. MacroMol., Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Env. & Life Sci., Okayama Univ.*, [3]*Dept. Biol. Chem. & Food Sci., Iwate Univ*)

L-Amino acid oxidase (LAO) is a flavoenzyme that catalyzes oxidative deamination of an L-amino acid to produce a 2-oxo acid with ammonia and hydrogen peroxide. Most of known LAO show low substrate specificity. Among them, LAO from Rhodococcus sp. AIU Z-35-1 (rzLAO) is found to exhibit broader substrate specificity than any other known LAOs. rzLAO is able to oxidize the ε-amino group of L-Lys, as well as the α amino group. Moreover, rzLAO can produce Nα-benzyloxycarbonyl-L-aminoadipate-σ-semi-aldehyde from Nα-benzyloxy-carbonyl-L-lysine. To understand the molecular mechanism of the unique substrate specificity of rzLAO, we purified and crystallized rzLAO. The rzLAO crystal was diffracted up to 2.1 Å resolution and the structure analysis is now underway.

2Pos009 Thermococcus kodakaraensis 由来エンドヌクレアーゼ NucS の構造
Structure prediction and analyses of endonuclease NucS from *Thermococcus kodakaraensis*

Setsu Nakae[1], Atsushi Hijikata[1], Toshiyuki Tsuji[1], Koki Yonezawa[1], Ken-ichi Kouyama[2], Kouta Mayanagi[3], Sonoko Ishino[4], Yoshizumi Ishino[4], Tsuyoshi Shirai[1] ([1]*Fac. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*, [2]*Grad. Sch. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*, [3]*Medical Inst. of Bioregulation, Kyushu Univ.*, [4]*Grad. Sch. Bioresource and Bioenviron. Sci., Kyushu Univ.*)

NucS is a novel endonuclease identified by screening of PCNA binding proteins from Archaea. Particular biological function and substrate specificity of NucS remain unclear. The knowledge-based modeling via supramolecular modeling pipeline suggested the similarity of NucS functional structure to that of type II restriction enzymes. We determined the structure of NucS from *Thermococcus kodakaraensis* in complex with DNA. As the result, the experimentally determined structures have verified the model prediction. Additionally, a knowledge-based model of NucS-PCNA-DNA complex was constructed based on the crystal structures, and a result of preliminarily analysis with EM was also consistent with the model prediction.

2Pos010 MARTINI 粗視化シミュレーションにおけるタンパク質の多段階構造変化手法の開発
Development of a method enabling multiple-step conformational change along a path in MARTINI coarse-grained simulations

Kaita Fujihara, Tatsuki Negami, Tohru Terada, Kentaro Shimizu (*Dept. of Biotech., Grad Sch. of Agri. Life Sci., Univ. of Tokyo*)

The coarse-grained molecular dynamics method is expected to be a powerful tool to investigate biologically relevant slow dynamics of a protein, such as conformational change. One of the most used coarse-grained models, MARTINI, is unable to change the protein structure during the molecular dynamics simulation because it must be used in combination with the elastic network mode to maintain the protein structure. In the present study, we developed a system enabling conformational change along a path by combining a multiple-basin potential method with the MARTINI force field. We implemented this system in Gromacs-5.0.2. We will discuss the effectiveness of this method in modeling a large-scale conformational change of a protein.

2Pos011 構造に分布を持つ二重スピンラベルタンパクの電子スピン共鳴の緩和および線形の解析
Relaxation and Lineshape Analysis for Electron Paramagnetic Resonance of Doubly Spin-labeled Protein with Structural Distribution

Yasunori Ohba[1], Munehito Arai[2], Jun Abe[3], Tetsuya Itabashi[1], Toshikazu Nakamura[3], Satoshi Takahashi[1], Seigo Yamauchi[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Art and Sci, Univ. Tokyo,*, [3]*IMS*)

Electron paramagnetic resonance (EPR) of a radical introduced in protein by the site-directed spin-labeling (SDSL) method is sensitive to local environment and dynamics of the aminoacid residue carrying the radical. By labeling protein at two aminiacid residues, further information is available from the spin-spin interaction between the radicals. We applied the double labeling method to the B domain of protein A (BDPA) to studie the relative dynamic motion between helices. The results showed discrepancy between the theoretical analysis and observation that may be accounted by distribution of structure which causes distribution of the inter-radical distance. In this paper, we show development of a method to analyze spin-spin interaction with distance distribution.

2Pos012 Structural insight into the interaction between Death-associated protein kinase 1 and natural flavonoids

Yuto Kosaka, Mineyuki Mizuguchi, Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

Death-associated protein kinase 1(DAPK1) is a Ca^{2+}/calmodulin dependent protein kinase. DAPK1 is a drug target of ischemic stroke and endometrial adenocarcinoma.
In the present study, we investigated the binding properties of DAPK1-natural flavonoids and identified that morin was the strongest binder among 17 selected flavonoids. To understand the structure-binding affinity relationship at the atomic level, we determined the DAPK1 crystal structures in complex with 8 selected flavonoids including morin at 1.4-2.0 Å. It was suggested the high affinity of morin was resulted from the ionic interaction between 2'-OH and K42. Otherwise, we found there are two binding modes in DAPK- flavonoids.
Binding characteristics of flavonoids will be discussed in detail.

2Pos013 ウシ心筋チトクロム酸化酵素 O 型中間体の X 線結晶構造解析

The X-ray structural analysis of O intermediate of bovine heart cytochrome *c* oxidase

Yuki Eto[1], Atsuhiro Shimada[1], Fumiyoshi Hara[2], Eiki Yamashita[2], Kyoko Shinzawa-Itoh[1], Tomitake Tsukihara[1,2], Shinya Yoshikawa[1] ([1]*Grad. Sch. Life Sci. Univ. Hyogo.*, [2]*Inst, Protein Res, Osaka Univ*)

Cytochrome *c* oxidase (CcO) pumps four protons coupled with sequential four electron transfers from cytochrome *c* to P intermediate giving F, O, E and R intermediates. But all four redox-active metal sites of CcO as prepared (Or) are in the oxidized state as in the O intermediate, Or is not involved in the enzyme catalytic cycle. For X-ray structural analysis, O intermediate crystals were prepared by incubation of reduced CcO crystals with O_2-saturated buffer. Regardless of the incubation time between 5 min and 90 min, X-ray structure shows various extents of detectable amount of the contaminant Or and R. Refinement of the X-ray structure obtained from the crystals prepared by 20 min incubation showing the lowest contaminant level is under way at 1.84 Å.

2Pos014 Molecular dynamics study of structural fluctuation in calmodulin

Hiromitsu Shimoyama, Mayuko Takeda-Shitaka (*Kitasato University, Faculty of Pharmacy*)

Calmodulin (CaM) is a calcium ion binding protein which mediates many essential biological processes. CaM is 148 amino acids long protein which forms approximately 7-8 α-helices in solution. The N- and C-terminal domains (NTD, CTD) are mutually connected by long linker-region, then, the overall CaM's structure is coarsely a symmetrical dumbbell-like shape. The ion binding to CaM causes conformation change. In order to understand the conformation change, structural fluctuations around holo-state was studied by molecular dynamics simulation.

2Pos015 タンパク質立体構造予測のための新規な疎水性指標

A novel measure for evaluating the satisfication of the hydrophobicity for protein structure prediction

Yota Masuyama, Koudai Takagi, George Chikenji (*Grad. Sch. Eng., Nagoya Univ.*)

Empirical energy function has played an important role in predicting protein structures. In these function, hydrophobic effect is often measured by accessible surface area (ASA), contact number (CN) or residue depth (RD), which reflects how buried each residue is in a protein. However, there is still much room for improvement in these measures. For example, it has been pointed out that ASA and RD cannot be used for course grained models and that CN is insensitive to evaluate residue environments. We, here, introduce a new measure evaluating hydrophobicity which is based on the CN but modified so as to be senstive to the environments. In this presentation, we will provide the detail description of the new measure and the results of the stucture prediction benchmark test.

2Pos016 アミロイド線維凝集に関する分子動力学計算を用いた静電相互作用の研究

Study of electrostatics interaction of amyloid fibrils using molecular dynamics calculation

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It became clear that KFFE which shortened KLVFFAE which is a part of amyloid β and these 7 residues make seat structure experimentally. However, we know that KFFK and EFFE which are structure like KFFE do not make seat structure by similar experiment.
In this study, it is assume that the difference is according to the electrostatic interaction. We simulate three pattern, single of KFFK, single of EFFE, mixture of KFFK and EFFE by molecular dynamic simulation. As a result, single simulation of KFFK and EFFE are not formed seat structure, but mixture simulation can confirm seat structure. These results agree with a laboratory finding.

2Pos017 α ヘリックスが β-ストランドペア形成に及ぼす影響

The effect of α-helices on β-strand pairing propensity

Hiromi Suzuki (*Sch. Agri., Meiji Univ.*)

We analyzed residue pairing patterns in β-sheets to understand the mechanism of β-sheet formation. We extracted PDB data less than 30% sequence identities and determined β-strand regions by DSSP. Protein domains were divided into two classes, mainly β and αβ domains according to the CATH criteria. Residue pairing propensities for anti-parallel pairs showed almost same between mainly β and αβ domains even if hydrogen-bonding (HB) and non-bonding (nHB) pairs and/or edge and central strands pairs were distinguished. On the other hands, 40 HB and 23 nHB pairs in parallel pairs showed different pairing propensity. These results may suggest that the presence of α-helices between β-strands has effect not only on forming parallel pairing but also on β-strand pairing propensity.

2Pos018 分子動力学シミュレーションを用いた Gads の SLP-76 認識機構の解析

Analysis of the SLP-76 ligand recognition mechanism of Gads by molecular dynamics simulation

Yoshiyuki Uemura, Kazuyoshi Ueda, Motoyasu Ozawa (*Yokohama National University*)

Intracellular signal transduction is an important mechanism for the growth and the differentiation of the immuno competent cell. Gads and SLP-76 are known as member protein such signal transduction pathway in cell. Meanwhile, SH3 is a domain which is known as one of the most abundant modules for protein interaction in nature, which can binds Pro-rich peptide strongly. Gads also includes a SH3 domain in its structure. However, SH3 domain in Gads does not recognize the Pro-rich peptide, instead, it recognizes the RxxK motif included in SLP-76. The mechanism of these specific binding nature has not been cleared yet. In this work, we tried to elucidate the binding mode and recognition mechanism between Gads and SLP-76 complex using the molecular dynamics simulation method.

2Pos019 Subunits kinetics in alpha-crystallin as studied by small angle neutron scattering

Rintaro Inoue[1], Takumi Takata[1], Norihiko Fujii[2], Nobuhiro Sato[1], Yojiro Oba[1], Shinichi Takata[3], Noriko Fujii[1], Masaaki Sugiyama[1] ([1]*Research Reactor Institute,Kyoto University*, [2]*Teikyo Univ, Radioisotope Res Ctr,*, [3]*Japan Atom Energy Agency*)

Alpha-crystallin is one of the major structural proteins in the eye lens. The most fascinating characteristic feature of α-crystallin is its chaperone activity, preventing the aggregation of various target proteins under external stresses such as heat and UV. One of the proposed concepts is the hybridization of α-crystallin with target proteins. Therefore it is strongly expected that α-crystallin possesses a dynamical structure, which is submitted to a constant reorganization following certain subunits kinetics. Monitoring such kinetics of subunits in α-crystallin must contribute to unveiling the unresolved mechanism of its chaperone activity. We utilized the combination of deuteration-labeling technique and time-resolved small-angle neutron scattering technique.

2Pos020 単粒子解析法を用いて 26S プロテアソームの立体構造解析
Structural analysis of the 26S proteasome by cryo-electron microscopy and Single-Particle Analysis

Zhuo Wang[1], Yasuo Okuma[1], Daiske Kasuya[2], Kaoru Mitsuoka[3], Yasushi Saeki[4], Takuo Yasunaga[1] ([1]*Department of Bioscience and Bioinformatics, Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [2]*Biomedicinal Information Research Center, Japan Biological Information Consortium (JBIC)*, [3]*Osaka University*, [4]*Laboratory of Protein Metabolism, Tokyo Metropolitan Institute of Medical Science*)

The 26S proteasome is composed of a barrel shaped 20S harbouring the proteolytic chamber together with one or two 19S. Since the weak assembly of the 26S makes structural analysis more difficult, the GraFix method was used to purify and stabilize holoenzyme 26S proteasome. After using GraFix, not only could the glycerol which was contributed to the low contrast of EM images be easy removed, but also the mount of particles could be increased. Even though the glycerol was removed, the EM images were still lacking in visibility. thus the Wiener filter which can remove the additive noise was used in here. Finally, we got high contrast images to pick up and obtained the 3D structures. After comparison, we consider that the 19S structure has been changed after losing Rpn10.

2Pos021 TRAF 結合分子の結晶構造と X 線小角散乱解析
Crystal structure and small-angle X-ray scattering analysis of TRAF binding protein

Teruya Nakamura[1], Chie Hashikawa[1], Yuya Yokote[1], Mami Chirifu[1], Yuu Taguchi[2], Jin Gohda[2], Taishin Akiyama[2], Kentaro Semba[3], Yoshinari Okamoto[1], Shinji Ikemizu[1], Masami Otsuka[1], Jun-ichiro Inoue[2], Yuriko Yamagata[1] ([1]*Grad. Sch. Pharmaceut. Sci., Kumamoto Univ.*, [2]*Inst. Medical Sci., Univ. Tokyo*, [3]*Dept. Life Sci. and Medical Bio-sci., Waseda Univ.*)

Tumor necrosis factor receptor (TNFR) associated factor (TRAF) proteins are major signal transducers involved in the TNFR superfamily and the Toll/interleukin-1 receptor family, and TRAF1~TRAF7 are identified in mammals. The C-terminal domain of TRAF1~TRAF6 participates in binding to receptors and adaptor proteins, and the E3 ligase activity of the N-terminal RING Zinc finger domain of TRAF2, TRAF3, TRAF5 and TRAF6 is crucial for the downstream signaling such as the activation of NF-κB and AP-1. In this work, we present the crystal structures and small-angle X-ray scattering data of the TRAF binding protein (TRAFBP), which is involved in the TRAF signaling pathway, and discuss the structural basis of the TRAF signaling mechanism through the interaction with TRAFBP.

2Pos022 緩和モード解析による転移温度付近の 10 残基のシニョリンのダイナミクス
Dynamics of 10-Residue Peptide, Chignolin near a Transition Temperature using Relaxation Mode Analysis

Ayori Mitsutake[1,2], Hiroshi Takano[1] ([1]*Dep. of Phys., Keio Univ.*, [2]*JST, Presto*)

We have developed relaxation mode analysis (RMA) to extract dynamic properties of proteins from long MD simulations. In RMA, slow relaxation modes and rates are extracted from MD simulations. Here, we apply RMA to extract good reaction coordinates for the system, in which there are large conformational changes such as folding/unfolding simulation. We performed a 750 ns simulation of chignolin near a transition temperature and observed many transitions between local-minimum-energy states. In RMA, we extract good reaction coordinates automatically. The free energy surfaces obtained by RMA give us more clear understanding of the transitions between local-minimum-energy states even though the simulation has large conformational changes.

2Pos023 大規模構造変化を許容するヘモグロビン結晶：結晶中タンパク質の動きの観測を目指して
Hemoglobin crystals that allow large-scale conformational changes: toward the observation of protein motions in crystals

Naoya Shibayama (*Div. of Biophysics, Jichi Medical Univ.*)

Many proteins undergo large-scale conformational changes that are relevant to their functions. The most studied example is the T (tense) to R (relaxed) transition in hemoglobin. Two crystal structures of the deoxy T and liganded R state have been well characterized, but the sequence and nature of the conformational transition linking these two end states are not known, which limits our understanding of the allosteric mechanism. We introduce a crystal form in which the protein is free to adopt any structure, depending on the conditions. A combination of such crystals and X-ray crystallography enables the observation of protein motions induced by changing external conditions. The reason why this crystal form allows large protein conformational changes will be discussed.

2Pos024 Circadian timing governed by cyanobacterial KaiC ATPase

Atsushi Mukaiyama[1,2], Jun Abe[1], Takuya Hiyama[1], Seyoung Son[3], Toshifumi Mori[2,4], Shinji Saito[2,4], Masato Osako[3], Julie Wolanin[1], Eiki Yamashita[5], Takao Kondo[3], Shuji Akiyama[1,2] ([1]*CIMoS, IMS*, [2]*Grad. Univ. for Adv. Studies, SOKENDAI*, [3]*Nagoya Univ.*, [4]*Dep. of Theor. and Comp. Molecular Science, IMS*, [5]*IPR, Osaka Univ.*)

KaiC is a core protein of the cyanobacterial circadian clock, and its ATPase activity exhibits circadian rhythm in the presence of two other clock proteins, KaiA and KaiB.
Recent studies propose that slow ATPase of KaiC is involved as a circadian pacemaker. To elucidate its molecular basis underlying slow ATPase, we examined the structure and function of KaiC. Key structural origins were obtained by our crystallographic analysis. In addition, we demonstrated that KaiC ATPase is under the control of intramolecular regulation. Relaxation kinetics by cancelling its regulation exhibits circadian timescale, and mutational analysis revealed that its speed is finely correlated with the frequency of the circadian clock. In this meeting, we discuss the details.

2Pos025 サチライシンにおける局所的な柔軟性増大のアロステリック効果

Allosteric effect of locally-enhanced flexibility in subtilisin

Takato Sato, Kosei Maetani, Dan Parkin, Jun Ohnuki, Koji Umezawa, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

Flexibility and allostery of proteins are both essential for function and regulation, and we focus on the relationship between flexibility and allostery. In serine protease subtilisin, mutants in which a β-turn is made flexible by glycine substitution increase their enzyme activity (Fuchita, et al. 2012). As this turn is distant from the catalytic site, there should be an allosteric effect caused by the locally-enhanced flexibility, the mechanism of which is unclear. By conducting molecular dynamics simulation, we found that the mutation softens surface loops and alters flexibility of key residues in the catalytic site. We discuss how this allostery is utilized in the catalytic function of this enzyme.

2Pos026 シナプス小胞膜を模倣した膜上でのαシヌクレイン線維形成メカニズム

Study on the mechanism of amyloidogenesis of α-synuclein on presynaptic membrane mimetics

Mayu Terakawa S.[1], Yuxi Lin[1], Tatsuya Ikenoue[1], Naoya Fukui[2], Yasushi Kawata[2], Young-Ho Li[1], Yuji Goto[1] ([1]*IPR, Osaka Univ.*, [2]*Dept. of Chem. and Biotech., Grad. Sch. of Eng., Tottori Univ.*)

Intrinsically disordered α-Synuclein (αSN) is a causative protein for Parkinson's disease due to its amyloid fibrillation. The binding of αSN to synaptic vesicles induces a conformational transition of the N-terminal region to α-helix; however, a role of helical states on fibrillation remains unclear. We herein examined the mechanism of αSN fibrillation depending on concentrations of lipids, a component of synaptic vesicles, using various spectroscopies. The fibrillation was inhibited at high concentration of lipids where αSN adopts helical structures whereas non-helical αSN disordered at low lipid concentrations increased amyloidogenicity. We propose that distinct initial structures of αSN in membrane-bound states play a key role for regulating amyloidogenesis of αSN.

2Pos027 アナモックス菌のラダラン脂質生合成にかかわると推定されるラジカル SAM 酵素の発現と精製

Expression and purification of a radical SAM enzyme, which is presumably involved in ladderane lipid biosynthesis in anammox bacteria

Jumpei Shimada[1], Nozomi Shinde[1], Tomoya Hino[1], Miki Fukuma[1], Ken Takai[2], **Shingo Nagano**[1] ([1]*Grad. Schl. Eng., Tottori Univ.*, [2]*JAMSTEC*)

Aanammox bacteria convert ammonium and nitrite into dinitrogen gas. The lipid membrane of anammox bacteria contains ladderane lipids, which has linearly concatenated cyclobutane ring. Comparative genomics suggest that a radical SAM enzyme (RS enzyme), which presumably contains FeS cluster, *S*-adenosyl methionine (SAM), and vitamin B_{12}, is involved in the the ladderane lipid biosynthesis. We expressed and purified the RS enzyme. A gene of the RS enzyme is amplified by PCR, using a metagenome from anammox membrane bioreactor or from geothermal water stream in Hishikari gold mine as a template. The gene is cloned into pET16b and transferred into C43(DE3) strain. Purified apo form of the RS enzyme was obtained by metal affinity and anion exchange column chromatography

2Pos028 揺らぎと水和効果に着目した ABC トランスポーターの NBD 二量体化過程の統計熱力学的解析

Statistical thermodynamic analysis of dimerization of nucleotide binding domains in an ABC transporter: fluctuation and hydration effects

Honami Sakaizawa, Hiroshi C. Watanabe, **Tadaomi Furuta**, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech*)

ATP-binding cassette (ABC) transporters are membrane proteins that transport substrates by ATP-driven dimerization of nucleotide binding domains (NBDs). However, the detailed mechanism of NBD dimerization remains unclear. Here, we investigated the driving forces for NBD dimerization of MJ0796 from the archaeon *Methanococcus jannaschii* by molecular dynamics (MD) simulations and three dimensional reference interaction site model (3D-RISM) theory. The free energy profile indicated that a long-range enthalpic interaction and a short-range entropic interaction mainly contribute the NBD dimerization. Furthermore, comparing the static and dynamic approaches, it was found that the thermal fluctuation is important for lowering the energy barrier between open and closed states.

2Pos029 区分同位体標識を用いた β_2-アドレナリン受容体のリン酸化の NMR 解析

NMR analyses of the phosphorylation of β_2-adrenergic receptor using segmental isotopic labeling

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Phosphorylation of the C termini of G-protein-coupled receptors (GPCRs) by GPCR kinases (GRKs) plays crucial roles in intracellular signal transduction. However, little is known about the phosphorylation sites and the conformation of the phosphorylated C termini of GPCRs. Here, we utilized NMR to investigate the structure of phosphorylated C terminus of β_2-adrenergic receptor (β_2AR). In order to overcome the signal overlaps, segmental isotopic labeling of the β_2AR C terminus was accomplished by protein *trans*-splicing. Our results revealed that several residues in the membrane-proximal region of β_2AR C terminus are phosphorylated by GRK2, and that C terminus undergoes conformational changes upon phosphorylation, which would be important for efficient signal transduction.

2Pos030 ホタルの発光に関与する酵素ルシフェラーゼの荷電性アミノ酸残基に対する滴定曲線の導出

Computational Analysis of Titration Curves of Some Amino Acid Residues in Proximity to Catalytic Center of Firefly Luciferase

Naohisa Wada[1], Keiici Horie[1], Sho Takamatsu[1], Itsuki Kaji[1], Hironori Sakai[2] ([1]*Fac. of Food Life Sciences, Toyo Univ.*, [2]*Insti. of Fluid Science, Tohoku Univ.*)

The enzymatic reaction of firefly bioluminescence is the oxygenation of luciferin (Ln) to excited-oxyluchiferin (Oxyln*) by luciferase (Luc). Oxyln* emits yellow-green or red light according to pH 7.8 or 6.8, respectively. In this study, the pKa's of some ionizable amino acid residues in proximity to Luc catalytic center were calculated basing on the crystallographic data (PDB:2D1R) by the Generalized Born method packaged in DS Ver.4.0 to find the characteristic difference from the experimental pKa's of the isolated corresponding amino acids in the aqueous solution. The main result shows calculated pKa's of some amino acids residues of Luc depend considerably on its initial structures to be relatively larger than those measured for the corresponding free amino acids.

2Pos031 阻害剤 BOF を結合したバクテリア XOR のリガンド結合ポケット入口に存在する一つのループの揺らぎについて

On the motion of a loop located in the entrance to the ligand-binding pocket of bacterial XOR with the inhibitor BOF

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BOF which is a structure-based inhibitor for xanthine oxidoreductase (XOR) fills most of the ligand-binding pocket of XOR, and strongly (weakly) inhibits the mammalian (bacterial) XOR as another inhibitor febuxostat. From 100ns MD simulations for the mammalian and the bacterial XOR with BOF, we found only in the case of the bacterial XOR that a loop located in the entrance to the ligand-binding pocket bends outward as if a gate opens. This motion of the loop might be significant to understand the binding or releasing mechanism of structure-based inhibitors. In this study, the loop opening motion from the results of our MD simulations on XOR with BOF is shown, compared with the results of our MD simulations on XOR with febuxostat, or on XOR itself.

2Pos032 等温滴定型熱量測定によるヒトヘモグロビンーIHP 間の相互作用の研究

Interactions between inositol hexakisphosphate and hemoglobin studied by isothermal titration calorimetry and oxygen binding measurements

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We have studied the interactions between inositol hexakisphosphate (IHP) and human tetrameric hemoglobin (Hb) -both in its liganded and unliganded forms- by isothermal titration calorimetry (ITC) and compared those results against oxygen binding measurements performed on native Hb, at pH 7.0 and 15 C. CyanmetHb and nickel-porphyrin Hb were used for the liganded and unliganded forms, respectively. ITC experiments showed that IHP binds to both derivatives, but neither Hb derivative isothermal titration curve could be fitted with a single, but rather a two binding site model. Oxygen binding experiments showed that IHP indeed binds to tetrameric liganded Hb lowering its oxygen affinity 10-fold.

2Pos033 3D-RISM 理論に基づく Pim1-リガンド系における結合自由エネルギーの予測

Estimation of binding free energy based on the 3D-RISM theory for the Pim1-ligand system

Takeshi Hasegawa[1], Masatake Sugita[1], Takeshi Kikuchi[1], Fumio Hirata[2] ([1]*Dept. of Bioinf., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Research Org. of Sci. and Tech., Ritsumeikan Univ.*)

To explore the drug candidate molecules, it is necessary to assess the binding free energy between a protein and a ligand. Although many computational methods for predicting the binding free energy has been proposed, these methods have some disadvantages, especially, evaluating the solvation thermodynamics of complicated solutes. The 3D-RISM theory can calculate the solvation free energy in a reasonable computational cost, although it can not be taken into account the structural fluctuation of a protein. In this study, we try to calculate the binding free energy combined MD simulations and the 3D-RISM theory. We apply this protocol to estimate binding free energy between Pim1 kinase and its inhibitor.

2Pos034 粗視化モデルを用いた T7 RNA ポリメラーゼの転写動態に関する考察

Study of transcriptional dynamics of T7 RNA polymerase using coarse-grained model

Kizuku Yamana, Hiraku Nishimori, Akinori Awazu (*Dept. of Math. and Life Sciences, Hiroshima Univ.*)

If DNA is damaged by radiation, DNA-protein cross-links (DPCs) damage, unusual covalent bond DNA and protein, often arise and stop the sliding of RNAP. Then, RNAP is congested by following RNAPs at damage spot. The recent experimental study showed that such congestions of T7 RNAP make frequent transcriptional errors.

We analyzed the mechanism of the errors caused by T7 RNAP congestions using coarse-grained molecular model. We obtained basic structure of a DNA, mRNA and T7 RNAP complex by Protein Data Bank (PDB code: 1msw) and construct the molecular models for following two situations, i) an isolated T7 RNAP interacts with DNA and binds a RNA as usual, and ii) T7 RNAPs are congested on DNA and their motions are influenced with each other.

2Pos035 蛋白質熱安定性に及ぼす共溶媒の添加効果

Effects of cosolvent addition on the thermal stability of a protein

Shota Murakami[1], Masahiro Kinoshita[2] ([1]*Grad. Sch. Energ. Sci., Kyoto Univ.*, [2]*Inst. Adv. Energ., Kyoto Univ.*)

It is experimentally known that addition of monohydric alcohol lowers the thermal stability of a protein and the degree of the lowering becomes higher as the size of its hydrophobic group increases, whereas addition of polyol possessing three or more hydroxyl groups enhances the stability and the enhancement becomes more conspicuous as the number of hydroxyl groups increases. We showed that a large gain in the translational, configurational entropy of water due to the reduction in water crowding in the bulk is the driving force of protein folding. Here we perform analyses on the effects of cosolvent addition, which are focused on the crowding effect. We investigate to what extent the experimental observations mentioned above can be elucidated by our theoretical method.

2Pos036 独立成分分析 tICA を用いたヒストンテールの遅い運動の解析

Slow dynamics of histone tails in molecular dynamics simulation revealed by time-structure based independent component analysis

Sotaro Fuchigami (*Grad. Sch. of Medical Life Science, Yokohama City Univ.*)

DNA transcription in eukaryotic cells is regulated by post-translational modifications of histone tails. It is considered that the flexibility of the histone tails is crucial for the transcription regulation, but details of their molecular mechanism remain unclear. In the present study, to characterize the flexible motion of the histone tails and to elucidate its underlying mechanisms, we performed MD simulations of histone tails of histone H2A/H2B dimer in explicit water. We focused on slow dynamics of histone tails and identified them by applying the time-structure based independent component analysis (tICA) to the simulation results. With additional analyses, the detected slow dynamics were confirmed to be actually occurred.

2Pos037 接着結合における張力伝達分子としてのβ-カテニン
β-catenin as a mechano-transmitter molecule at adherens junctions

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At intercellular adherens junction (AJ), cells transmit force with each other and stabilize the balance, contributing to force-driven mechanisms in multicellular dynamics. Here, we focused on β-catenin, a mechano-transmitter protein at AJ, and analyzed the mechanical behaviors by employing atomic force microscopy (AFM). As a result, force versus extension curves showed that β-catenin behave as a nonlinear elastic material under tension, i.e., a mechanical component with low stiffness under low tension and with high stiffness under high tension. Such behaviors suggest mechanical functions of β-catenin as a mechano-transmitter at AJ: 1) preserving links between AJ molecules under low tension and 2) transmitting high tension with high mechanical stability.

2Pos038 Spectroscopic and calorimetric analysis for conformational stability of c-Myb DNA-binding domain under different pH conditions

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The effects of pH on conformational stability of the minimum DNA-binding domain of c-Myb, Repeat 2 and 3 (R2R3) are assessed under different pH conditions, ranging from 4.0 to 7.5. The secondary structural analysis showed that the helical content is significantly decreased, while thermal stability is increased at mildly acidic pH. In order to investigate the pH-dependent conformational change in more detail, two dimensional NMR measurements were performed, and showed that most crosspeaks corresponding to the R2 region were selectively disappeared as pH decreased until 5.0, indicating that R2 exists as a partially disordered state. The large chemical shift changes were also observed on the crosspeaks corresponding to the R3 region between N-terminus and to second helix.

2Pos039 高速時間分割蛍光異方性によるタンパク質間相互作用の解析
Protein-protein interaction revealed by time-resolved fluorescence anisotropy

Akane Kato[1], Etsuko Nishimoto[2] ([1]*Grad. Sch. Bioresour. Bioenviron. Sci., Univ. Kyushu*, [2]*Fac. Agr., Univ. Kyushu*)

Precise description for weak protein-protein interaction has not been established. In this study, we extracted experimental parameters concerning with the interaction from the rotational diffusion of protein. Time-resolved fluorescence anisotropy, which could discriminate the entire rotation of the protein from the fast segmental motion, was employed. The interaction parameter, k_{rot} estimated from time-resolved measurements, showed that the larger attractive interaction would be induced by NaCl than that from results of steady-state studies. This difference is due to the contribution from the fast segmental motion inevitably included in steady-state anisotropy. To characterize protein-protein interaction *in vivo*, we will extract new parameters in concentrated solution.

2Pos040 高圧小角散乱法により検出されたニトリラーゼ会合体の高圧中間体解析
Analysis on pressure intermediate of Nitrilase oligomer detected by high-pressure small-angle scattering

Tetsuro Fujisawa[1,2,3], Ryo Ishiguro[1,2] ([1]*Dep. Eng., Gifu Univ.*, [2]*Harima Inst., Riken*, [3]*NSSR, Nagoya Univ.*)

Nitrilase is an industrial enzyme that hydrolyzes nitrile compounds into ammonia and carboxylic acids. Since this enzyme detoxifies cyanoproducts to be hydrolyzed, nitrilase superfamilly is widely spread among various species. The microbial nitrilase associates from an inactive dimer to an active oligomer upon heating. This activation with oligomerization is reported to be common in nitrilase superfamilly. We have extensively examined this homo-oligomerization with high pressure and found that the pressure reversal of oligomerization consists of two stages. We analyzed scattering data under various conditions by global fitting procedure.

2Pos041 フェリチンアセンブリ反応における静電相互作用の重要性
Importance of electrostatic interactions during ferritin assembly reaction

Daisuke Sato, Atsushi Kurobe, Satsuki Takebe, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. Bioinfo., Grad. Sch. Eng., Soka Univ.*)

Ferritin (Ftn) is a spherical shell-shaped protein with 24 subunits. Ftn dissociates into dimers at acidic pH and reassembles into the native structure when pH increases. To study importance of electrostatic interactions in Ftn assembly, we did the stopped-flow time-resolved small angle X-ray scattering experiments. The assembly rate depended on pH and ionic strength. This indicates that there are repulsive electrostatic interactions between dimers. A possible interaction is the repulsion between net charges of dimers. If this is true, the assembly rate is expected to increase with reducing net charges. To test this possibility, we made several mutants with different net charges. The result agreed well with this expectation and showed importance of net charge repulsion.

2Pos042 アクチンフィラメントの圧電特性 II
Piezoelectric property of an actin filament II

Jun Ohnuki[1], Takato Sato[1], Taro Q.P. Uyeda[2], Mitsunori Takano[1] ([1]*Dept. of Pure & Appl. Phys., Waseda Univ.*, [2]*Biomedical Res. Inst., AIST*)

Various cell function is regulated by external force. Although, in molecular level, proteins are thought to serve as mechanosensor, it remains unclear how proteins sense and transduce external force. Therefore, we have investigated the response of the mechanosensing proteins, myosin and actin filaments, to applied force by conducting molecular dynamics (MD) simulation, finding that myosin shows piezoelectric effect: the applied force redistributes the electric charges on the surface of myosin. In contrast, we could not clarify such an electrostatic response in an actin filament at a high confidence level (Annual meeting, 2014). Thus, we improved the reliability of our MD simulation and here ascertain whether piezoelectricity is also inherent in actin filaments.

2Pos043 天然変性領域である p53C 末端ドメインの構造と物性にアセチル化が及ぼす影響

Effect of acetylation on structure and physicochemical properties of the intrinsically disordered C-terminal domain of p53

Shinji Iida[1,2], Hironobu Hojo[1], Haruki Nakamura[1], Junichi Higo[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*)

The C-terminal domain (CTD) of p53 is an intrinsically disordered region. Although it is known that acetylation of CTD influences the affinity with its interaction partner molecules, their physicochemical behavior has not yet understood well. Moreover, it is still unclear how acetylation affects the physicochemical properties of CTD. To reveal this effect, we did all-atom virtual-system coupled multicanonical molecular dynamics (V-McMD) simulations and circular dichroism (CD) measurements on each of the non-acetylated and acetylated CTD systems. While the spectra from the CD measurements do not differ significantly, a free energy landscape computed from the V-McMD simulations shows that the acetylation influences the structural properties largely.

2Pos044 天然タンパク質の分子密度に関する統計解析

Statistical analysis on the molecular density of native proteins

Hidenobu Kawai, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

The molecular density of a protein is a basic and important property that represents the extent of stiffness and packing of a protein. However, the relationship between the molecular density and the structure and function of proteins is poorly understood. Here, we calculated the densities of 39,451 protein domains classified in the SCOPe database. We found that small proteins with disulfide bonds and/or ion binding have high densities, while the α/β-type structures were less observed in the highly dense proteins. In terms of functional classification based on superfamily, toxins are the most frequent in the highly dense proteins. These results indicate that the molecular density of native proteins is closely related to proteins' structure and function.

2Pos045 ALS を引き起こす SOD1 の線維形成に伴う物性変化

Physical property change associated with fibril formation of ALS-causative SOD1

Noriko Fujiwara[1], Michiru Wagatsuma[2], Daisaku Yoshihara[1], Yoshiaki Furukawa[3], Haruhiko Sakiyama[1], Hironobu Eguchi[1], Keiichiro Suzuki[1] ([1]*Dep. of Biochem., Hyogo coll. of Med.*, [2]*Biosensor, Res. and Dev., ULVAC*, [3]*Dept. of Chem., Keio Univ.*)

Cu/Zn-superoxide dismutase (SOD1) catalyzes the conversion of superoxide anion radical into less reactive molecular oxygen and hydrogen peroxide, thereby protecting cells against oxidative stress. On the other hand, mutations of SOD1 gene cause familial amyotrophic lateral sclerosis (ALS) characterized by formation of abnormal SOD1-immunopositive inclusion bodies in the spinal cord. Missfolding of SOD1 results in fibrils, soluble oligomers and amorphous aggregates, which are thought to be associated with ALS. We found that wild-type SOD1 undergoes to hydrogelation accompanied by the formation of amyloid-like fibrils. We will report the dynamic process of hydrogelation of SOD1 fibrils monitored by QCM technic.

2Pos046 構造エレメントの決定因子としての分子内相互作用

Intramolecular interaction as a determination factor for structure elements of proteins

Yuji Konno, Takeshi Inagaki, Hironari Kamikubo, Mikio Kataoka (*Graduate School of Materials Science, Nara Institute of Science and Technology*)

We have proposed that a protein consists of structure elements and linkers. We assume that the structure elements are kernels of intramolecular interactions to maintain a tertiary structure. To examine the hypothesis, we have newly developed software to evaluate the intramolecular interaction strengths. Intramolecular interaction is evaluated by contact volume which is defined by the overlapped volume within van der Waals contacts. The software enables to predict structure elements through the contact volume which is calculated by the crystal structure coordinates from Protein Data Bank (PDB). We evaluate the software performance by calculating the contact volumes of staphylococcus nuclease and will compare them with the structural elements determined experimentally.

2Pos047 示差走査型蛍光定量法を用いたアルドケト還元酵素阻害剤の選択性の評価

Evaluation of selectivity of aldo-keto reductase inhibitors using differential scanning fluorimetry

Yuji O. Kamatari[1], Aurangazeb Kabir[2], Satoshi Endo[3], Kazuo Kuwata[2] ([1]*Life Sci. Res. Center, Gifu Univ.*, [2]*United Grad. Sch. Drug Discov. Med. Info. Sci., Gifu Univ.*, [3]*Lab. Biochem., Gifu Pharm. Univ.*)

Inhibitors of AKR1B10 belonging to the aldo-keto reductase (AKR) superfamily are considered to be the promising candidates of anti-cancer drugs. Although AKR1B1 is structurally similar isoform of AKR1B10, it involves in glucose metabolism. Thus selective inhibition of AKR1B10 is required for the development of the anti-cancer drugs. In this study, we firstly compared correlation between T_m and IC_{50}, obtained from the differential scanning fluorimetry (DSF) and enzyme inhibitory experiment, respectively, and found a good correlation between them excluding compounds with low-solubility. Then, we estimated their selectivity as an inhibitor against five major human AKR family proteins and found that the DSF method is quite useful for developing specific AKR1B10 Inhibitors.

2Pos048 GlcNAc 含有高分子を用いたビメンチンの新たな機能の解明

Elucidation of novel functions of vimentin with the well-defined GlcNAc-bearing polymers

Sadanori Yamasaki[1], Horihiko Ise[2], Tadashi Nakamura[1], Yoshiko Miura[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Univ. Kyushu*, [2]*IMCE., Univ. Kyushu*)

Recently, the development of molecular-targeting reagents is focused for effective cancer therapies. We have reported that cytoskeletal protein, vimentin, possesses a N-acetylglucosamine (GlcNAc)-binding property especially on cell surface. Vimentin has reported to highly express in various types of tumors and thus expected as a candidate of new therapeutic markers. In this study, we examined vimentin behaviors such as cell-surface recruitment and GlcNAc-binding property, employing the molecular-weight well-defined GlcNAc-bearing polymers synthesized by living radical polymerization. The polymers were found to be able to interact with HeLa cells via vimentin on the cell surface. Effect of chain length of the polymer on its vimentin binding activity is reported.

2Pos049 Protective effect of model peptides for group-3 LEA proteins on enzymes during desiccation

Takao Furuki, Minoru Sakurai (*Center for Biological Resources and Informatics, Tokyo Institute of Technology*)

The two short peptides were tested as candidates as protective reagents for dried enzymes. One is referred to as PvLEA-22 made of two tandem repeats of the 11-mer motif characteristic to group-3 late embryogenesis abundant (G3LEA) proteins originated in African sleeping chironomide, and the other is PvLEA-44 with such four tandem repeats. When lactate dehydrogenase (LDH) was dried alone, its enzyme activity was virtually lost. However, the LDH activity was maintained more than 70 % when either PvLEA-22 or PvLEA-44 was added to LDH before drying. Furthermore, the cooperative protective effects were observed when trehalose was also added to LDH together with such short peptides before drying. Thus, these model peptides are promising protective reagents for dried enzymes.

2Pos050 NMR と分子動力学シミュレーションによるラン藻由来アルカン合成酵素の構造ダイナミクス解析
Structural dynamics of a cyanobacterial alkane-producing enzyme, aldehyde deformylating oxygenase, studied by NMR and MD simulations

Yuma Suematsu, Yuuki Hayashi, Munehito Arai (*Dept. Life Sci., Univ. Tokyo*)

Aldehyde deformylating oxygenase (AD) is a key enzyme for alkane biosynthesis in cyanobacteria. Crystal structure of AD has shown that it is composed of eight α-helices and binds two iron atoms. Interestingly, its substrate, a fatty aldehyde, binds inside of the barrel-shaped AD protein. However, the intrinsic dynamic properties and catalytic motions of AD remain unclear. To elucidate the structural dynamics of AD, we have constructed the *E. coli* expression system of AD from *Thermosynechococcus elongatus* BP-1, purified the protein, and measured the NMR spectra. We have also performed molecular dynamics (MD) simulations of AD and found that the most dynamic region in AD is located close to the putative substrate-entry site.

2Pos051 四量体型サルコシン酸化酵素の分子動力学シミュレーション :生成物の輸送経路の解明
Molecular dynamics simulation of heterotetrameric sarcosine oxidase: analysis of channeling of product

Daisuke Nakajima[1], Go Watanabe[2], Haruo Suzuki[2], Shigetaka Yoneda[2] ([1]*Grad. Sch. Sci., Kitasato Univ.*, [2]*Sch. Sci., Kitasato Univ.*)

Heterotetrameric sarcosine oxidase (HSO) catalyzes the oxidation of sarcosine to form glycine and hydrogen peroxide. The water channels within HSO through which substrates and products can migrate have been determined by the probability density distribution of water molecules in our previous molecular dynamics simulations. The umbrella sampling method was applied to the motion of the iminium intermediate product in the sarcosine-reduced form of HSO in the present study. The selective transport pathway was discussed using the free energy.

2Pos052 生体高分子中性子結晶構造解析における水素の高感度検出のためのラジカル分子のタンパク質単結晶への導入
An introduction of radical molecules into a protein single crystal for more sensitive detection of hydrogen in neutron crystallography

Naoya Komatsuzaki[1], Takahiro Iwata[2], Yoshiyuki Miyachi[2], Toshiyuki Chatake[3], Katsuhiro Kusaka[4], Nobuo Niimura[4], Ichiro Tanaka[4,5] ([1]*Grad. Sch. of Sci. and Eng., Ibaraki Univ.*, [2]*Faculty of Sci., Yamagata Univ.*, [3]*RRI, Kyoto Univ*, [4]*Frontier Res. Center*, [5]*Coll. of Eng., Ibaraki Univ.*)

The technique of proton polarization in a protein single crystal is expected to improve the detection sensitivity of hydrogen by approximately eight times in comparison with conventional neutron protein crystallography. This technique is realized by dynamic nuclear polarization, which requires a sample crystal to be randomly doped by stable paramagnetic materials up to the ratio of 1 : 1000 = radical : hydrogen approximately, and so on. In order to realize and confirm the above condition, X-ray crystallography and ESR (Electron Spin Resonance) method were employed for a single crystal of Lysozyme protein doped by TEMPOL. As relatively positive results were obtained, the details will be discussed in the presentation.

2Pos053 Raf の生細胞内構造分布を明らかにする 1 分子 FRET 計測
Single-molecule FRET measurement to investigate structural distribution of Raf in living cells

Kenji Okamoto[1], Kayo Hibino[2], Yasushi Sako[1] ([1]*RIKEN*, [2]*NIG*)

Single-molecule fluorescence imaging has been successfully used to investigate dynamics of biomolecules in or on living cells. Those experiments often aim molecules on plasma membrane since total internal reflection illumination, for suppression of background signals, can excite only fluorophores near the glass surface and cameras cannot capture molecules freely diffusing in cytoplasm. Here we introduce the single-molecule FRET measurement for molecules diffusing deep inside a living cell. A FRET histogram is constructed from fluorescence bursts detected from single molecules passing through the focus spot of a confocal microscope. The system is applied to investigate the intracellular structural distribution of Raf and its change upon EGF stimulation of cell.

2Pos054 On the convergence of binding free-energy landscape calculation by improved virtual-system coupled adaptive umbrella sampling

Bhaskar Dasgupta, Junichi Higo, Haruki Nakamura (*Institute for Protein Research, Osaka University*)

A variant of adaptive umbrella simulation (AUS) “virtual-system coupled AUS” (V-AUS) enhances sampling along a virtual degree of freedom and calculates binding free-energy landscape along a reaction coordinate [Higo et al, J. Comput. Chem. in press (2015)]. The V-AUS quickly converges than the conventional AUS. Here, further improvements of V-AUS in all-atom receptor-ligand binding in water are presented. First, an initial set of conformations is generated by putting ligands in different grids around receptor. Second, sampling errors are removed by Markov approximation. Third, a set of novel fitting schemes parameterizes the biasing force in terms of reaction coordinate. We conclude that by using such advanced set of methods larger biological systems are tractable.

2Pos055 X 線 1 分子追跡法によるミオシン分子構造揺らぎの高精度リアルタイム計測

Molecular fluctuation analysis of myosin by DXT measurement

Keigo Ikezaki[1], Hiroshi Sekiguchi[2], Naoto Yagi[2], Toshio Yanagida[3], Yuji C. Sasaki[1,2] ([1]*Tokyo Univ.*, [2]*Spring-8/JASRI*, [3]*RIKEN/QBiC*)

Many researchers have investigated and revealed their kinetics and structural information using various kinds of bulk and single molecule observations. Further, it is important to investigate the state of molecular surface for better understanding to functions of proteins. However, it has not been clarified because of lacks of temporal and special resolutions of conventional single molecule observations. To tackle this problem, we has been trying to apply the novel high accuracy observation; Diffracted X-ray Tracking (DXT*), to reveal structural fluctuation of single myosin molecule.
(*)DXT method allows us to know the orientations of gold nano-crystals attached to protein surfaces with high special and temporal resolution. (SPring-8,BL40XU)

2Pos056 フェリチンからの鉄イオン遊離に与える交流磁場の影響

Iron exit from ferritin under alternating magnetic fields

Yuta Yamada, Tsuyoshi Hondou, **Hidetake Miyata** (*Facuty of Science, Tohoku University*)

We have shown that 50 Hz magnetic fields enhance DNA double strand break in fibroblast cells (Nakayama et al., 2012). For deeper understanding of this phenomenon, we examined if the magnetic field alters the exit of iron ions from iron storage protein, ferritin. Ferritin solution was exposed to 3.2 mT sinusoidal magnetic fields (50 Hz or 1 kHz) for 2.5 h or 5 h, and iron ions were quantified with an iron indicator ferrozine. Change in the absorbance at 562 nm was quantitatively analyzed with a two-step kinetic model: in the first step iron ion translocates from the interior to the external surface of ferritin, which is followed by the release of the ion into surrounding milieu. Only at 50 Hz, 2.5 h exposure the first step was slightly accelerated.

2Pos057 1 分子光子計数データのデータ同化解析

Sequential data assimilation for single-molecule FRET photon-counting data

Yasuhiro Matsunaga[1], Yuji Sugita[1,2,3,4] ([1]*RIKEN AICS*, [2]*RIKEN*, [3]*RIKEN iTHES*, [4]*RIKEN QBiC*)

Data assimilation is a statistical method designed to improve the quality of numerical simulations in combination with real observations. We have developed a sequential data assimilation method that incorporates one-dimensional time-series data of single-molecule FRET (smFRET) photon-counting into conformational ensembles of biomolecules derived from "replicated" molecular dynamics (MD) simulations. A particle filter using a large number of replicated MD simulations with a likelihood function for smFRET photon-counting data is employed to screen the conformational ensembles that match the experimental data. In the poster, we show the details of our method as well as preliminary results obtained by an application to the smFRET data of protein folding dynamics.

2Pos058 がん関連サイトカインに結合するラクダ科単一ドメイン抗体の試験管内淘汰と、その簡便・迅速なプルダウン法

***In vitro* selection and a simple and rapid pull-down assay of Camelidae single-domain antibodies that bind to cancer cytokine**

Hidenao Arai, Mana Mizugai, Koji Matsuoka, Naoto Nemoto (*Grad. Sch. Sci. Eng., Saitama Univ.*)

Single-domain antibodies (sdAb) are promising biomolecules for molecular recognition as compared with the conventional antibodies such as immune-globulin G (IgG) because of its thermostability and the ease preparation by bacterial culture. In this study, we have identified three kinds of Camelidae against a cancer cytokine by cDNA display method, that is one of genotype-phenotype linking technology involving with the evolutionally protein engineering. These sdAbs have been shown to have a binding ability for the cancer cytokine by a novel pull-down assay using a puromycin linker. We will discuss the usefulness of an *in vitro* selection system combining “cDNA display” for selection with the easy pull-down assay for rapid evaluation.

2Pos059 α ヘリカルタンパク質の設計

Design of diverse all-α protein strucrures based on αα-rule

Kouya Sakuma[1,2], Rie Koga[1], Nobuyasu Koga[1,2,3] ([1]*CIMoS, IMS.*, [2]*SOUKENDAI*, [3]*JST, PRESTO*)

α-helical proteins previously designed de novo have been limited to relatively simple ones such as helical bundle or coiled-coil. In this project, we aim to design all-α proteins with more complex topologies like the globin-fold. As for α and β proteins, we previously reported rules on the junctions between adjacent secondary structure elements, namely ββ, βα and αβ-rule, and designed ideal proteins with various topologies. Here, we extended the rules by investigating α-α junctions in naturally occurring protein structures; geometries of α-α-units can be characterized by the connecting loop types. We applied the rules to generate diverse all-α folds from scratch.

2Pos060 An attempt to improve the method for directed evolution by in vitro compartmentalization

Kentaro Tahara[1], Ryo Iizuka[1], Ayaka Iguchi[2], Hyun Yoon Dong[2], Tetsushi Sekiguchi[2], Shuichi Shoji[2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Grad. Sch. of Adv. Sci. and Eng., Waseda Univ.*)

In vitro compartmentalization (IVC) is a powerful technique for directed evolution of proteins using cell-like compartments such as emulsions and liposomes. In each compartment, a single-copy gene is transcribed and translated to link the gene and the gene product. It has been difficult to screen compartments containing proteins with desired activities and to recover the genes. To overcome the problems, we envisioned an improved IVC method using microbeads displaying multiple copies of single genes, which were prepared by single-molecule emulsion PCR. Using this method, we tried to acquire the genes encoding fluorescent proteins from libraries of the GFP gene including random sequences in the chromophore region.

2Pos061 Incorporation of a photoisomerizable non-natural amino acid into proteins for photo-control of protein functions

Rumi Shiba, Takayoshi Watanabe, Takahiro Hohsaka (*School of Materials Science, Japan Advanced Institute of Science and Technology*)

Incorporation of azobenzene, a photoisomerizable compound which undergoes cis-trans isomerization, into proteins makes it possible to control protein functions by irradiation. However, usual chemical modification of proteins with azobenzene compounds is non-site-specific and cannot be available for this purpose. To achieve site-specific incorporation of azobenzene into proteins, azobenzene-containing non-natural amino acid, AzoAla, was incorporated in response to amber codon.Here, we applied this strategy to cell-free synthesis of a photo-controllable single-chain antibody. To increase the yield of AzoAla-containing proteins, the incorporation of AzoAla was also investigated in E. coli using AzoAla-specific aminoacyl-tRNA synthetase mutant and an amber suppressor tRNA.

2Pos062 癌抑制タンパク質 101F6 のアスコルビン酸からの電子受容機構

Electron transfer mechanism from ascorbate to human tumor suppressor 101F6 protein

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Human tumor suppressor 101F6 protein belongs to cytochrome b561 family and is known to induce caspase-independent apoptosis in cultured cancer cells by utilizing redox reactions involving AsA. Our previous mutation analyses on 101F6 protein by conferring putative AsA-binding site of neural b561 indicated that mutants containing F67Y mutation had spectral characters similar to those of neural b561. Further, DEPC-treatment on these F67Y mutants caused significant inhibition of AsA-dependent electron transfer but not for others. We analyzed DEPC-modification sites by MALDI-TOF-MS analysis. Results suggested that hydrogen bonds formed by the Tyr residue near the cytosolic heme might have important roles for the electron-transfer from AsA for neural b561 but not for 101F6.

2Pos063 ハーフメトヘモグロビン M を用いたヒト成人ヘモグロビンの酸素親和性制御に関する α 鎖と β 鎖の Fe-His 結合の役割の違いに関する研究

Study on different roles of Fe-His bond between α and β chains for oxygen affinity regulation of human hemoglobin by using half-met Hb Ms

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Half-met hemoglobin Ms (Hb M), in which heme iron of either α or β chain is occupied by Fe^{3+} instead of Fe^{2+}, are very excellent specimens to elucidate discriminately a regulation mechanism of oxygen affinity (OA) of α or β normal chain, since Hb Ms as valence-hybrid ($Fe^{2+}Fe^{3+}$) Hb can bind O_2 to Fe^{2+} of normal chain only. In Hb M Iwate (αH87Y) and Hb M Boston (αH58Y), which form $\alpha Fe^{3+}\beta Fe^{2+}$, Fe-His stretching frequencies increased as OAs of β subunit rose. In contrast, those of Hb Saskatoon (βH63Y) and Hb M Milwaukee (βV67E), which form $\alpha Fe^{2+}\beta Fe^{3+}$, did not show pH dependence in spite of pH dependence of OA and exhibited at least two components of Fe-His band. We discuss a relationship among OA, Fe-His frequency of normal chain and quaternary structure of each Hb M.

2Pos064 酸素センサー酵素 YddV のヘム結合グロビンドメインの結晶構造解析

Crystal structure of isolated heme-bound globin domain of a heme-based oxygen-sensor enzyme, YddV, from *Escherichia coli*

Jotaro Igarashi, Toru Kikuchi, Ariki Matsuoka (*Dept. Nat. Sci., Biol., Sch. of Med., Fukushima Med. Univ.*)

YddV from *Escherichia coli* is a novel heme-based oxygen-sensor enzyme with diguanylate cyclase (DGC) activity that synthesizes c-di-GMP from two molecules of GTP. The YddV protein is composed of a heme-bound globin domain in the N-terminal region (YddV-heme) and a functional domain with the DGC active site in the C-terminal portion. In the present study, The structure of Fe(III) form of YddV-heme was solved by multiple anomalous dispersion methods, based on the heme iron absorption edge. YddV-heme forms a dimer. Heme distal residues are hydrophobic; including Leu65, Met69, and Phe42, and there is no water molecule in the heme pocket. Tyr43 are pointed out of the heme pocket.

2Pos065 チトクロム酸化酵素に対するチトクローム c の 2 つの結合部位の X 線構造解析

X-ray structural analysis of two binding sites of cytochrome *c* to cytochrome *c* oxidase

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Cytochrome *c* oxidase (C*c*O) reduces molecular oxygen, coupled with proton pumping using electrons from cytochrome *c* (Cyt.*c*). For elucidation of the mechanism of electron transfer from Cyt.*c* to C*c*O, it is desirable to determine the crystal structure of the Cyt.*c*-C*c*O complex at high resolution. We have succeeded in obtaining the two types of complex crystals, Cyt.*c*/C*c*O = 1.0 and Cyt.*c*/C*c*O =1.5. The former crystals diffracted X-ray up to 1.8 Å. The calculated density map revealed the presence of Cyt.*c* bound to near the Cu_A of C*c*O and the electron transfer pathway from Cyt.*c* to C*c*O. In the latter crystals, the second Cyt.*c* was observed near the subunit Ⅳ and Ⅷ. This is one of the strongest evidence in support of the existence of two binding sites of different affinity.

2Pos066 三量体ハロロドプシンにおけるカロテノイド結合能の特異性

Specificity of carotenoid binding to halorhodopsin trimer

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A light driven anion pump halorhodopsin (NpHR) exists on the biomembrane of *N. pharaonis*. The trimeric NpHR binds a kind of carotenoid, bacterioruberin (BR), at the hydrophobic interface.
In this study, we investigated the carotenoid-binding specificity of the trimer NpHR obtained from a heterologous over-expression system in *E. coli*.
Although several types of carotenoid (β-carotene, astaxanthin and lycopene) were mixed to the trimer NpHR solution, these did not bind to the NpHR at all.
In addition, acetylation of BR at its hydroxyl groups also decreased the affinity to the NpHR.
These results indicate that the binding specificity of the NpHR to the BR is very high.

2Pos067 脂質平面膜法を用いた TRPM3 チャネルの機能解析
Single channel analysis of the TRPM3 channels in planar lipid bilayers

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TRPM3 is a non-selective cation channel and activated by nifedipine and neurosteroid, pregnenolone sulfate (PS). TRPM3 is mainly expressed in nervous systems, and reported to be involved in heat sensation. However, the functional characterization of TRPM3 channels still remain poorly understood. To investigate the single channel properties of TRPM3, we analyzed the purified TRPM3 protein in planar lipid bilayers. Application of nifedipine caused increase in open probability. Unlike nifedipine, application of PS alone did not induce the channel openings and co-application of the PI(4,5)P2 was required. In addition, we found that increase in temperature did not induce the channel openings, indicating that TRPM3 is unlikely to represent a temperature sensor by itself.

2Pos068 分子シミュレーションによる阻害剤が結合した ADP/ATP carrier の内向き開構造
Structure of the inhibitor-bound inward-facing conformation of ADP/ATP carrier: A simulatioin study

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ADP/ATP carrier is a transporter protein embedded in mitochondrial inner membrane. Its functional role is to exchange cytosolic ADP for ATP in matrix space. Although X-ray structure representing outward-facing state has been available, inward-facing structure is not known. Utilizing a recently developed enhanced sampling method, linear response path following (LRPF), we have revealed an atomic structure for the inward-facing conformation of ADP/ATP carrier. Furthermore, the structure enabled us to search for the inhibitor-bound conformation of the protein. The atomic structure of the protein-inhibitor complex revealed the role of aliphatic chain of the inhibitor in trapping the protein in the inward-facing state. Details of the structure will be discussed.

2Pos069 ケイジド化合物導入による低分子量 G タンパク質 H-Ras の多量体形成の光制御
Photo-control of small G-protein H-Ras multimerization using caged compounds

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Recently we found that the chemical modification of the cysteine residues in the C-terminal hyper variable domain (HVR) of small G protein H-Ras with hydrophobic thiol reactive reagents induces multimer formation of H-Ras. Utilizing the phenomenon, we tried to control the transition between the monomer and multimer of H-Ras. We employed thiol reactive caged-compound, 2-Nitrobenzyl bromide (NBB). The modification of H-Ras with NBB induced formation of multimer. Light irradiation at 400 nm liberate the 2-Nitrobenzyl group resulted in the multimer changed over to monomers. It is suggested that modification of the cysteines in HVR with caged compound enable to photo-control the multimerization of Ras.

2Pos070 細菌外膜シトクロムにおける代謝共役したフレキシブルな構造と機能
Flexible Strucutral Alteration Associated with Respriatory Electron Flow in Microbial Outer Membrane c-type Cytochrome Complex

Akihiro Okamoto, Yoshihide Tokunou, Chinotaikul Punthira, Kazuhito Hashimoto (*Grad. Sch. Eng., Univ. Tokyo*)

Specific microbes conserve energy by transporting electrons from cell interior electron chain to exterior insoluble electron acceptors, a process referred to as extracellular electron transport (EET). We recently found the kinetics of EET process is strongly dependent on the formation of semiquinone active species bound to outer-membrane c-type cytochromes (OM c-Cyts), which operate as not only electron but also proton carrier. Here, our in-vivo and in-vitro analysis shows that the OM c-Cyts flexibly alters their strucutre for stablizing bound flavin cofactor depending on the rate of upstream respiratory electron flow. This finding imply that OM c-Cyts has the function to maintain intracellular redox homeostasis in accordance with bacterial metabolic activity.

2Pos071 結晶化を目指したカイコガ性フェロモン生合成活性化神経ペプチド受容体（PBANR）のアンタゴニストの探索
Ligand screening of silkmoth pheromone biosynthesis-activating neuropeptide receptor for crystallization of the ligand-receptor complex

Yukie Katayama[1], Hidekazu Katayama[2], Takeshi Kawai[1], Tatsuya Suzuki[1], Tatsuki Ebisawa[1], Ryo Natsume[3], Yu-Hua Lo[4], Toshiya Senda[4], Toshihiro Nagamine[5], Masaaki Kurihara[5], Jae Min Lee[5], Joe J. Hull[6], Shogo Matsumoto[5], Hiromichi Nagasawa[1], Koji Nagata[1], Masaru Tanokura[1] ([1]*UTokyo*, [2]*Tokai Univ.*, [3]*TDU*, [4]*KEK-PF*, [5]*RIKEN*, [6]*USDA-ARS*)

The silkmoth pheromone biosynthesis-activating neuropeptide receptor (PBANR), a class-A GPCR, regulates the biosynthesis of sex pheromone by interacting with the 33-a.a. peptide ligand PBAN. We obtained a low resolution X-ray diffraction image (12 Å resolution) for the PBAN-PBANR complex, where the third intercellular loop of PBANR was mostly replaced by T4 lysozyme (T4L) for stabilization. PBANR is unstable without its ligand after solubilization from the cell membranes. We have been screening the ligand(s) of PBANR suitable for the stabilization and crystallization of the ligand-receptor complex. We will present our in vitro binding data of some ligand candidates and PBANR, and compare the in vitro data with in vivo activity data of these compounds.

2Pos072 チャネル電流計測を用いた脂質二重膜中における L-, D-ボンビニンの抗菌活性の解析
Antimicrobial activity analysis of L- and D-Bombinin in lipid bilayers using channel current recording

Yusuke Sekiya[1], Hirokazu Watanabe[1], Yuki Kitahashi[2], Izuru Kawamura[2], Ryuji Kawano[1] ([1]*Tokyo Univ Agr Tech*, [2]*Grad. Sch. Eng, Yokohama Nattl Univ.*)

Bombinins are antimicrobial peptides which make the stereoisomers. They have an L- or D-amino acid residue at the 2nd position from the N-terminus, called bombinin H2 (L) and bombinin H4 (D). Only the difference of the chirality at the single amino acid causes the clear distinction of the antimicrobial activities (AMAs). Moreover, it has known that H2 and H4 mix at 1:1 has the highest AMAs. Though extensive studies have spectroscopically been conducted, these mechanisms have still been unclear. In this study, we used bacteria model cell-membranes prepared by a microdroplet system for analyzing the AMAs in terms of transmembrane current signals. We believe this analysis will be helpful for creating more effective therapeutic agents for infections with molecular basis.

2Pos073 Active supercomplex purified from bovine heart reveals the functional unit of the mitochondrial respiratory chain

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To understand the roles of mitochondrial respiratory chain supercomplexes, methods for consistently preparing supercomplexes must be established. Amphipol-stabilized supercomplex purified from bovine heart did contain complexes Ⅰ, Ⅲ, and Ⅳ at a ratio of 1:2:1; six molecules of Q_{10}; and 623 atoms of phosphorus. When cytochrome *c* was added, the supercomplex exhibited KCN-sensitive NADH oxidation. We measured the resonance Raman spectrum of the reduced amphipol-solubilized supercomplex and the mixture of amphipol-solubilized complexes $Ⅰ_1$, $Ⅲ_2$, and $Ⅳ_1$ using an excitation wavelength of 441.6 nm, allowing measurement precision comparable to that obtained for complex Ⅳ alone. The purified active sample may provide insights into the effects of supercomplex formation.

2Pos074 上皮成長因子受容体（EGFR）の膜貫通および膜近傍ドメインの構造ダイナミクス解析
Conformational dynamics of transmembrane and juxtamembrane domains of epidermal growth factor receptor (EGFR)

Ryo Maeda[1], Takeshi Sato[2], Kenji Okamoto[1], Takehiko Inaba[3], Yasushi Sako[1] ([1]*Cellular Informatics Lab., RIKEN*, [2]*Inst. for Protein Research, Osaka Univ.*, [3]*Lipid Biology Lab., RIKEN*)

The transmembrane (TM) helix and associated juxtamembrane (JM) domains bridge the extracellular and intracellular domains of single-pass membrane proteins including EGFR, and play an important role for molecular activation. While the JM region interacting with negatively charged lipid molecules such as PIP2 regulates EGFR activation, coupling between dynamics of TM and JM remains unclear. Here, by using synthesized fluorescent peptides consist of TM and JM (TM-JM) regions, it is designed to elucidate the dynamics of TM and JM domains and the interaction of JM domain with lipid molecules. These peptides were reconstituted into nanodisc, a small membrane particle, and applied to single-molecule FRET analysis to monitor the conformational dynamics of TM-JM regions.

2Pos075 Characterization of RuvA-RuvB-Holliday junction DNA complex formation using Zero Mode Waveguides

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The Escherichia coli RuvA-RuvB protein complex promotes branch migration of Holliday junction DNA. RuvA is a Holliday junction DNA specific DNA binding protein and interacts with RuvB. RuvB is an ATPase that belongs to the AAA+ protein class and forms hexameric ring in an ATP-dependent manner. To characterize RuvA-RuvB-Holliday junction DNA complex in more detail, in this study, we constructed Cy5-labeled RuvB and counted the number of the RuvBs binding to a RuvA-Holliday junction DNA using Zero Mode Waveguides. Our data demonstrated that the numbers of Cy5-RuvBs binding to a RuvA-Holliday junction DNA increased in order of no nucleotide, with ADP, with ATPγS, with ADP and ATPγS.

2Pos076 The stability of the translation arrest of SecM internal deletion mutants

Yuanfang Guo, Zhuohao Yang, Ryo Iizuka, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

SecM, a bacterial secretion monitor protein, contains a specific amino acid sequence at its C-terminus, called arrest sequence, which interacts with the ribosomal tunnel and arrests its own translation. Although the arrest sequence is sufficient and necessary for translation arrest, we have recently found that the nascent SecM chain outside the ribosome stabilizes the translation arrest. To identify the region responsible for the stabilization, SecM internal deletion mutants fused to the C-terminus of HaloTag via a glycine-serine linker were expressed in an in vitro transcription-translation system, and their lifetimes of the translation arrest were measured. In the meeting, the details of the results will be discussed.

2Pos077 高活性 TALEN の高活性化機構の解析
In vitro analysis of super-active TALEN

Yoko Terahara[1], Kazuho Ikeda[1], Naoyuki Miyasita[2], Yasushi Okada[1] ([1]*QBic, Riken*, [2]*CDB, Riken*)

Gene editing in vivo has become possible by the development of artificial nucleases that can be designed to cut the genome DNA selectively at the target site. TALENs are highly specific artificial nucleases. However in mammalian cells and embryos, TALENs often show poor activity. Recently we have developed “super-active” TALEN and demonstrated that which can mediate efficient genome editing in mouse embryos.
To evaluate how our “super-active” mutations actually affect enzymatic properties, we have produced active TALEN proteins and measured their activities in vitro. In addition, we investigated the structural differences between super-active and conventional TALE. Our in vitro based approaches provide us a new insight for further improvement of TALEN techniques.

2Pos078 弾性ネットワークモデルによる DNA の配列・構造・運動と機能の関係の考察
Analysis of the relationship among sequences, structures, motions and the functions of DNA by elastic network models

Shuhei Isami[1], Sayuri Tatemoto[1], Atsushi Ikegaya[1], Hiraku Nishimori[1,2], Naoaki Sakamoto[1,2], Akinori Awazu[1,2] ([1]*Dept. Math. and Life Sciences, Hiroshima Univ.*, [2]*Research Center for Mathematics on Chromatin Live Dynamics*)

Construction of the elastic network models of DNA and the normal mode analysis of them are performed in order to reveal the structure-dynamics relationships for several nucleotide sequences. First, we propose the all atom elastic network model of DNA that can explain the profiles of temperature factors in several crystal structures of DNA. Second, we propose a coarse grained elastic network model of DNA described only by the C1’ carbon coordinates in sugar chain, that reproduce the nucleotide sequence dependent dynamics obtained by the all atom elastic network model. Using this model, we clarify the general features of sequence-dependent structure-dynamics relationships of DNA with the length similar to those of nucleosome forming regions.

2Pos079 DNA 二重鎖をほどく１分子計測とシミュレーションによる水素結合の構造へのエントロピー寄与

Entropic effects of hydrogen bonds to the double-stranded DNA structure revealed by single-molecule mechanical unzipping

Akihiro Fukagawa[1], Michio Hiroshima[2], **Makio Tokunaga**[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*RIKEN*)

To elucidate the role of hydrogen bonds in DNA structure, we carried out single-molecule mechanical unzipping of DNA using intermolecular force microscopy. We have resolved DNA unzipping forces into single hydrogen-bond forces. The rupture energy per single hydrogen bonds to be 1 to 2 k_BT, which is almost the same as the thermal energy. Molecular dynamics simulations of mechanical unzipping showed that the force curves had a good agreement with the experiments. Entropy and Gibbs free energy were examined in MD simulations as functions of the number of formed hydrogen bonds, and it was found that they showed an unexpected feature in a different manner between GC and AT pairs. This result suggests that entropy plays an important role in the stabilization of DNA structure.

2Pos080 磁場によるプラスミド DNA への影響

The effect of magnetic fields on the plasmid DNA

Masahiko Hasegawa, Tsuyoshi Hondou, Hidetake Miyata (*Department of Physics, Tohoku University Graduate School*)

Epidemiological studies have reported that children living near the power line were at a risk of infant leukemia. Because infant leukemia occurs the result of genetic abnormalities, we focused on a possible hydroxyl radical-mediated damage of DNA, previous study has reported that hydroxyl radical produced by Fenton-like reaction damaged DNA and the damage was enhanced by magnetic field. In this study, hydroxyl radical was produced by horseradish peroxidase (HRP) and H_2O_2. Plasmid was exposed to or not exposed to magnetic fields. The plasmid damage was assessed by electrophoresis. In order to quantitatively evaluate the result, we made a plug-in analyzer running on ImageJ. We will discuss the effect of magnetic field exposure on plasmid integrity.

2Pos081 DNA 高次構造転移における３価陽イオンと２価陽イオンの競合効果

Mutual inhibition between 2+ and 3+ cations to induce folding transition of DNA

Chika Tongu[1], Takahiro Kenmotsu[1], Yuko Yoshikawa[2], Kenichi Yoshikawa[1] ([1]*Doshisha Univ.*, [2]*Ritsumeikan Univ.*)

We have performed single DNA observation on the higher order structure of giant DNA molecules in solution in the presence of cation with different valency by fluorescence microscope. We discovered divalent cations such as Mg(2+) and Ca(2+) induces loose shrinkage. On the other hand, SPD(3+) causes compaction on the giant DNA. Interestingly, it is found that divalent cations induce unfolding of the compact DNA in the presence of SPD(3+). Such antagonistic effect between 2+ and 3+ cations will be well explained by taking into account of the effect of the change in translational entropy of counter cations.

2Pos082 熱ショックタンパク質の発現調節をする温度感知 RNA の分子動力学シミュレーション

Molecular dynamics simulations of a temperature-sensitive RNA that regulates the gene expression of heat-shock proteins

Yoshiharu Mori[1], Hisashi Okumura[1,2] ([1]*IMS*, [2]*SOKENDAI*)

Non-coding RNAs are known to have important functions for many molecular biological processes. Some non-coding RNAs regulate gene expression. One example is a bacterial messenger RNA (mRNA) that is translated into heat-shock proteins. While a ribosome binding site of the mRNA molecule forms a secondary structure under normal conditions, the base pairs of the binding site are destroyed under high-temperature conditions. Ribosomes can bind the mRNA molecule by the base-pair breaking at high temperature. We studied the heat-induced structure changes of the mRNA using molecular dynamics simulations. We will discuss the thermoregulation mechanism related to its mRNA.

2Pos083 DNA および酵素のハイドロゲルカプセル内封入による単純なセルオートマトンの化学実装に向けて

Toward chemically implementing simple cellular automaton by encapsulating DNA and Enzyme in hydrogel capsule

Ibuki Kawamata[1], Satoru Yoshizawa[1], Fumi Takabatake[1], Ken Sugawara[2], Satoshi Murata[1] ([1]*Tohoku University*, [2]*Tohoku Gakuin University*)

We propose a theoretical model and discuss some challenging issues of experimental implementation of chemical reaction network using DNA and enzyme encapsulated in hydrogel capsule. By numerically solving partial differential equations that represent complex reaction-diffusion system of DNA bistable system, we have succeeded in simulating a spatio-temporal evolution that solves a simple maze problem. The key idea to solve the problem is arranging hydrogel capsules in a lattice, which makes it possible for the bistable system to perform a state transition rule of a simple cellular automaton. Hopefully, the framework to design such larger scale DNA computing will be applied to control molecular robots.

2Pos084 DNA のよじれ合いと巻き付きにおけるカイラリティの選択性

Chiral Selection in Braiding and Wrapping of Double-Stranded DNA

Sosuke Sano[1], Tomohiro Yanao[1], Kenichi Yoshikawa[2] ([1]*Grad. Sch. Sci. & Eng., Waseda Univ.*, [2]*Grad. Sch. Life & Med. Sci., Doshisha Univ.*)

Braiding and wrapping of DNA are of fundamental importance in replication, gene regulation, and chromatin packaging. This study highlights the selectivity of chirality in braiding and wrapping of DNA. We develop a coarse-grained model of B-form DNA, which consists of two elastic chains that mutually intertwine in a right-handed manner forming a double-stranded helix. Monte Carlo simulations of this model suggest that two juxtaposed DNA molecules can exhibit a spontaneous tendency to braid in a left-handed manner due to their asymmetric elasticity between bending and writhing. The DNA model also exhibits a chiral selectivity to wrap around a core particle in a left-handed manner, indicating the physical origin of the uniform left-handed wrapping of DNA in nucleosomes.

2Pos085 オーダーN法第一原理分子動力学計算のデモンストレーション

Demonstration of order-N first-principles density functional theory-molecular dynamics calculations

Takao Otsuka[1], Michiaki Arita[2], Makoto Taiji[1], David R. Bowler[3], Tsuyoshi Miyazaki[4] ([1]*RIKEN QBiC*, [2]*Grad. Sch. Sci. and Tech., Tokyo Univ. of Sci.*, [3]*UCL*, [4]*NIMS*)

Molecular simulation methods are now commonly used to explore biological phenomena of proteins and/or biomolecules. Such molecular simulation methods are expected to help us understand the mechanism of biological phenomena. Recently, quantum mechanics/molecular mechanics (QM/MM) hybrid methods or its molecular dynamics (QM/MM-MD) method are also used well in this field. Very recently, we succeeded to introduce MD method to our order-N first-principles density functional theory (DFT) method, called CONQUEST program. Our method is corresponding to full QM. In this poster, we will show the performance of our order-N QM-MD calculations and the preliminary results of our on-going study on hydrated DNA system.

2Pos086 一般化ボルンモデルにおける塩橋の安定性

Salt bridge stability in the generalised Born model

Dan Parkin, Yukinobu Mizuhara, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

The electrostatic interaction stabilizes protein and complex structures. The salt-bridges (SB) are important, but the estimation of its stability is not easy due to subtle balance between the electrostatic interaction and the Born energy, causing a serious problem in molecular dynamics (MD) simulations with the generalized Born (GB) model. It is known that this problem can be resolved by adjusting the intrinsic radius, which roughly represents the atomic radius, even though the underlying physical mechanism is unclear. By conducting MD simulations of a SB-forming simple system, we show that the intrinsic radius breaks the balance between the Born and electrostatic interaction energies when atoms become closer, providing the physical basis of how the SB in stabilized.

2Pos087 剛体球参照系を用いた密度汎関数理論に基づく新しい溶媒和自由エネルギー表式

A Solvation-Free-Energy Functional: A Reference-Modified Density Functional Formulation

Yutaka Maruyama[1], Tomonari Sumi[2], Ayori Mitsutake[3] ([1]*AICS, RIKEN*, [2]*Dep. Chem., Okayama Univ.*, [3]*Dep. Phys., Keio Univ.*)

The solvation-free-energy (SFE) is regarded as one of the most important properties for understanding the thermodynamics stability, including for protein folding and protein-ligand binding. The three-dimensional reference interaction site model (3D-RISM) theory, which is an integral equation theory for molecular liquids, overestimates the absolute values of SFE for large solute molecules in water. To improve the free-energy density functional for the SFE, we propose a reference-modified density functional theory (RMDFT), which employs hard-sphere fluids as there reference system. We show that using RMDFT can significantly improve the absolute values of the SFE for a set of neutral amino acid-side chain analogues as well as for a mini-protein chignolin.

2Pos088 MM/3D-RISM 法を用いた 2-Hydroxypropyl-β-Cyclodextrin の抱接現象における結合自由エネルギーの解析

Analyzing the binding free energy for the inclusion process of 2-Hydroxypropyl-β-Cyclodextrin by means of the MM/3D-RISM method

Masatake Sugita[1], Fumio Hirata[2] ([1]*Dept. of Bioinfo., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Research Org. of Sci. and Tech., Ritsumeikan Univ.*)

The calculation of the binding free energy requires evaluation of the configuration integral. This requirement makes the prediction of free energy by the MD simulation difficult. The 3D-RISM theory avoids the difficulty by integrating the solvent configuration analytically. In this study, we develop a protocol to predict the binding free energy in combination of a MD simulation with the 3D-RISM theory. As a benchmark test, we analyze an inclusion process of some small molecules by 2-Hydroxypropyl-β-Cyclodextrin. We try to predict the binding mode by the umbrella sampling, and generate the ensemble of the complex structure using MD simulation. After that, we calculate the binding free energy based on the MM force field and the 3D-RISM theory.

2Pos089 機械的な力による上皮形態形成制御

Mechanical control of epithelial morphogenesis

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We will present our ongoing projects on mechanical control of epithelial tissue morphogenesis.
1. Insect wings in natural populations exhibit astonishing morphological diversity. Recently, we developed a unified formalism that quantifies forces/stresses and tissue and cell kinematics in a developing tissue at an unprecedented quantitative level. The method will prove a basis of comprehensive analyses of the tissue shape determination.
2. We seek to identify the molecular mechanism whereby force-generating and force-sensing properties of F-actin and actin binding proteins regulate cell rearrangement dynamics and shape a tissue.

2Pos090 血管新生における内皮細胞動態ライブイメージング

Live cell imaging of vascular endothelial cell dynamics in angiogenesis

Naoko Takubo[1], Kazuaki Naemura[1], Ryo Yoshida[2], Terumasa Tokunaga[3], Osamu Hirose[4], Yasunobu Uchijima[1], Yukiko Kurihara[1], Hiroki Kurihara[1] ([1]*Graduate School of Medicine, The University of Tokyo*, [2]*The Institute of Statistical Mathematics, Research Organization of Information and Systems*, [3]*Faculty of Computer Science and Systems Engineering, Kyushu Institute of Technology*, [4]*College of Science and Engineering, Kanazawa University*)

Angiogenesis is a complex process accomplished by sprouting, elongation and bifurcation. Recently, we reported an angiogenic morphogenesis driven by collective endothelial cell (EC) movement called cell mixing, in which ECs move backwards and forwards, overtaking each other even at the tip [1]. However, individual and collective EC behaviors remain unknown. In this study, we developed a cell tracking system to delineate complex movement of ECs. Moreover, we demonstrated time-lapse live imaging for in vitro angiogenic model based on mouse aortic ring assay using the cell tracking system. As a result, individual EC dynamics in cell mixing (e.g., overtaking, overlapping and moving backward) were quantitatively evaluated.
[1] S. Arima, et al., Development. 2011;138:4763-76.

2Pos091 胎生期脳組織に発現するメカノトランスダクション関連遺伝子の解析

The functional analysis of the mechanotransduction-related gene expressed in the developing brain

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Previous studies showed that substrate stiffness acted as the extrinsic factor for stem cell fate determination in vitro. However, the molecular mechanism referred to as mechanotransduction is still unclear in vivo.
To determine this mechanism, we analyzed the developing brain as a model because of its spatiotemporal shifts in stiffness. First, we performed the in situ hybridization database search for identifying the molecules, which respond to stiffness. According to searching, we found a candidate gene associated with vinculin and actin. It is strongly expressed in the intermediate zone, which exhibits the highest stiffness in the developing brain. Here we report the histological analysis and gain of function analysis of this molecule in the developing mouse brain.

2Pos092 水中におけるアクチン重合の統計熱力学について

On the statistical thermodynamics of actin polymerization in aqueous solution

Tomohiko Hayashi[1], Hiraku Oshima[1], Makoto Suzuki[2], Masahiro Kinoshita[1] ([1]*Inst. Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Eng., Tohoku Univ.*)

Oosawa presented a simple theory of statistical mechanics of actin polymerization in the equilibrium state (*J. theor. Biol.* **1970**, *27*, 69). This theory is based on the postulation that a decrease in the system free energy occurs upon binding of an actin monomer to the end of an actin polymer, but the physical origin of this decrease is not clear. Here we quantify this free-energy decrease and decompose it into thermodynamically insightful components using our statistical-mechanical method. The effects of hydration as well as those of protein-protein direct interactions are fully taken into account. Further, we calculate the change in the partial molar volume upon actin polymerization and its temperature dependence. The effects of salt addition are also analyzed.

2Pos093 ESR 距離測定による心筋トロポニン I と T のリン酸化調節の 動的構造基盤

Structural dynamics of cardiac troponin regulated by phosphorylation, as studied by distance measurement using spin-labeling dipolar EPR

Hiroaki Yamashita[1], Chenchao Zhao[1], Koichi Sakai[1], **Keisuke Ueda**[1], Tatsuhito Matsuo[2], Satoru Fujiwara[2], Shoji Ueki[3], Toshiaki Arata[1] ([1]*Dept. Biol., Sci. Grad. Sch., Osaka Univ.*, [2]*JAEA*, [3]*Tokushima-Bunri Univ.*)

Heart muscle beating is regulated or abnormally modulated by phosphorylation or disease-causing mutagenesis of cardiac troponin (cTn) I and T. We have determined the secondary structure of N-terminal phosphorylatable extension of cTnI by measuring the distance distribution between two spin labels attached on i and i+4 residues. Upon binding with cTnC, PKA- and PKC-phosphorylatable regions formed more stable conformations whereas the other regions still exhibited unstable conformations. In preliminary study, we found that the distance distribution between cTnT and tropomyosin became narrower upon being docked on actin filament. This will be a framework to examine the effect of PKC phosphorylation and disease-causing mutagenesis on the cTnT-tropomyosin interaction.

2Pos094 ミオシンフィラメント懸濁液の ADP 存在・非存在下でのプロトン NMR 緩和経過

Spin-spin relaxation of 1H NMR signals from myosin filaments suspension with or without ADP

Tetsuo Ohno, Toshiko Yamazawa (*Department of Physiology The Jikei University School of Medicine*)

The dynamic changes of water molecules structure surrounding contractile proteins might play an important role in cross-bridge cycling during contraction. The spin-spin relaxation process of 1H-NMR signals from suspension of myosin filaments prepared from rabbit could be well represented by the summation of several exponentials indicating that water molecules in the suspension could be conveniently grouped into several components based on the relaxation time constant (T2). The slowest two components (T2 around 0.4s and 0.15s) dominated over faster relaxation components. This may suggest that the potential of the water molecules existing around myosin filaments is high.

2Pos095 OH 伸縮振動のラマン分光によるミオシン S1 の水和状態の測定

Hydration study on myosin subfragment-1 (S1) by Raman OH-stretching spectroscopy

Yuki Ochiai[1], Hideyuki Ohsugi[1], George Mogami[1], Tetsuo Taniuchi[2], Makoto Suzuki[1] ([1]*Grad. Sch. Eng., Univ. Tohoku*, [2]**FRIS., Univ. Tohoku*)

To understand the motor mechanism of myosin, changes in the structure and hydration states of myosin subfragment-1 (S1) intermediates during the chemo-mechanical cycle have been the important issue to be resolved. In this study, Raman OH-stretching spectroscopy was used to analyze the hydration states of S1 at 20℃. OH-stretching band of S1 solution was decomposed into those of solvents at 20℃ and different temperature. A criterion which required the total spectrum to be composed of fundamental vibration bands was adopted to extract the hydration water component. As a result, about three times larger hydration numbers were obtained compared to the previous DRS result ((Suzuki et al., Biophys J 1997,72, 18-23)).

2Pos096 光学顕微鏡および放射光 X 線回折実験による甲虫飛翔筋の構造的考察

Structural insights in coleopteran flight muscles by optical microscopy and synchrotron X-ray diffraction measurements

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Insects change the direction and the speed during flight while keeping the flapping motions at 20 - 1000 Hz. Recently, Sato's group found that one of the flight muscles, the wing folding muscle (WFM), can steer flying beetle. Their finding leads us to consider that each type of insect flight muscles has its unique role in the flight. Here, we report sarcomere lengths, myofilament lengths and filament lattice spacing in different kinds of flight muscles in a beetle, Mecynorrhina torquata, determined by using optical microscopes and synchrotron X-ray diffraction. We found that the sarcomere of WFM was about 1.5 times longer than dorsal longitudinal muscle (DLM). We will discuss how the sarcomere structures relate to the physiological roles of each flight muscle.

2Pos097 Mg/Ca 結合 F-アクチンの多重水和状態
Multi-hydration States of F-actin Bound with Mg/Ca Ions

Ryotaro Chishima[1], Asato Imao[1], George Mogami[1], Takahiro Watanabe[1], Tetsuichi Wazawa[2], Nobuyuki Morimoto[1], Makoto Suzuki[1,3] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Inst. Sci. Ind. Res., Osaka Univ.*, [3]*FRIS, Tohoku Univ.*)

Multi-hydration states of actin were revealed by dielectric relaxation (DR) spectroscopy. G- and F-actin in Ca- or Mg-contained HEPES buffers exhibited dual hydration components consisting of restrained water with DR frequency f2 (< fw : of pure water) and hypermobile water (HMW) with DR frequency f1 (>fw). Hydration state of F-actin strongly depended on the ionic composition. Restrained water numbers could be explained as the number of water molecules locating mostly in the first water layer of the protein atomic structures. Number of HMW molecules was roughly explained by the volume between the equipotential surface of -kT/2e and the first water layer around the actin surface by solving the Poisson-Boltzmann equation using CHIMERA (UCSF).

2Pos098 In-plane 2D buckling of microtubule under compressive stress

Arif Md. Rashedul Kabir[1], Daisuke Inoue[1], Kazuki Sada[1,2], **Akira Kakugo**[1,2] ([1]*Fac.of Sci., Hokkaido Univ.*, [2]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*)

Microtubules (MTs) in cell are often found in buckled state with short wavelength, whereas isolated MTs buckle at long arcs in vitro. This difference in buckling behavior has been attributed to the surrounding elastic medium in cell as supported by the 'elastic foundation model', although no experimental evidence has been available yet. Here, we investigated the role of elastic crosslinker in MT buckling. We show that, crosslinker plays the key role in determining the buckling mode of MTs. Comparison of our experimental results with 'elastic foundation model' suggests that interaction between MT and crosslinker should be considered in depicting the buckling behavior of MT.

2Pos099 Emergence of ultra-large vortex pattern by collectively moving microtubules on application of external indentation stimuli

Tamanna Ishrat Farhana[1], Daisuke Inoue[2], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Fac. of Sci., Hokkaido Univ.*)

The emergence of different type of patterns due to the self-organization of collectively moving microtubules is a fascinating phenomenon. The application of controlled external mechanical stress on self-organized microtubules causes formation of different patterns. The application of single step and cyclic stretching stimuli re-orient the dynamic microtubules network into more ordered alignment along the perpendicular of the stretching axis and in zigzag fashion respectively. While an ultra large vortex pattern forms by applying indentation stress. The effect of extent of indentation on the dimension and radius of curvature of the vortex with time is also investigated.

2Pos100 ATP 存在下のミオシン S1 はアクチンフィラメントのらせんピッチを伸ばしコフィリン結合を阻害する：高速 AFM による直接観察
Active myosin S1 induces longer helical pitches of actin filaments and inhibits actin binding of cofilin as demonstrated by high speed AFM

Kien Ngo[1], Noriyuki Kodera[2], Taro Uyeda[1] ([1]*Biomed. Res. Inst., AIST*, [2]*BioAFM FRC., Kanazawa U.*)

We previously showed biochemically that binding of the motor domain of myosin II (S1) cooperatively changes the structure of actin filaments and allosterically inhibits cofilin binding. Here, we used high speed AFM to image real-time binding of cofilin to actin filaments immobilized on positively charged lipid surface in the presence or absence of S1. We found that transient binding of S1 in the presence of a low concentration of ATP strongly inhibits cofilin binding, decreases filaments with shorter helical pitch and increases filaments with significantly longer helical pitch. This active S1-induced untwisting of actin filaments presumably plays crucial roles in inhibition of cofilin binding since cofilin prefers to bind to actin filaments with shorter helical pitch.

2Pos101 バクテリア由来セルラーゼ *Cellulomonas fimi* Cel6B の逐次運動の１分子蛍光可視化解析
Single-molecule fluorescence imaging analysis of processive movement of a bacterial cellulase *Cellulomonas fimi* Cel6B

Daiki Ishiwata[1,2,3], Akihiko Nakamura[1,2,3], Tomoyuki Tasaki[4], Ryota Iino[1,2,3] ([1]*Okazaki Inst. for Integrative Bio.*, [2]*Inst. for Molecular Science*, [3]*Sch., Phis. Sci., SOKENDAI*, [4]*Sch. Eng., Univ. Tokyo*)

We have recently shown that a fungal cellulase *Trichoderma reesei* Cel6A (TrCel6A) is a linear molecular motor which processively hydrolyzes and moves on crystalline cellulose. This study aims to assess if a bacterial cellulase *Cellulomonas fimi* Cel6B (CfCel6B) is also a linear molecular motor. From a crystal structure of TrCel6A and a modeling of CfCel6B structure, TrCel6A and CfCel6B seem to have different loop structures around the catalytic site which affect the processivity. A cysteine mutant of CfCel6B was generated, expressed in *E. coli*, purified and labeled with Cy3. We confirmed processive movements of CfCel6B on cellulose Iα by single-molecule fluorescence imaging. Average velocity was 13.5 nm/s (N = 36 molecules) and larger than that of TrCel6A (~8 nm/s).

2Pos102 Individual kinesin immobilization on microfabricated nano-pillar arrays

Taikopaul Kaneko[1], Fumie Oda[1], Takahide Kon[3], Ryuji Yokokawa[2] ([1]*Grad. Sch. Eng., Univ. Kyoto*, [2]*Grad. Sch. Eng., Assoc. Prof, Univ. Kyoto*, [3]*Grad. Sch. Eng., Dep. Biol. Sci., Prof, Univ. Osaka*)

In the eukaryotic mitosis, chromosomes are duplicated and equally distributed to two daughter cells. Choromokinesin, a member of kinesin-10 family, involve in prometaphase chromosome congression to spindle equator. Such polar ejection force generated by multiple motors has not been measured. Toward its measurement, we established a platform to immobilize kinesins with the defined spacing, which was realized by fabricating nano-pillar arrays. Pillar diameter and spacing was designed as 50 nm and 100 nm, respectively. Distance between kinesins was measured by tracking a short microtubules, and showed good agreement with the designed pillar spacing. Our result could help to measure a force produced by multiple motors in mitosis.

2Pos103 細菌Ⅲ型分泌機構の解明を目指したエフェクター輸送のリアルタイム評価系の構築

Real-time evaluation of effector transport for clarifying the transport mechanism on type III secretion apparatus

Takashi Ohgita, Tsubasa Uekawa, Kyoko Momiyama, Naoki Hayashi, Naomasa Gotoh, Kentaro Kogure (*Kyoto Pharm. Univ.*)

Like drug-injection, bacteria inject effector proteins, related to infection or toxicity, into host cells via needle-like type III secretion apparatus (T3SA). However, the mechanism of effector transport through T3SA is unclear. Previously, we found out that T3SA rotates on proton-motive force and it relates to effector transport. It was also reported that conserved-aromatic residue in needle-component align helically in T3SA needle. Based on these, we hypothesized that T3SA might secrete effectors by rotating opposite direction to the helix. If this is true, the rotation- and secretion-speed of T3SA would correlate linearly. To confirm this, we attempted to construct real-time evaluation system for effector transport. In this presentation, we would report the results.

2Pos104 電場印加によるアクトミオシンの運動速度の可逆的変化

A reversible change in the velocity of the motility of actin filaments on a heavy meromyosin-coated surface under an electric field

Kuniyuki Hatori[1], Taiki Abe[1], Reito Wada[2] ([1]*Grad. Sch. Sci. Eng., Yamagata Univ.*, [2]*Grad. Sch. Med., Yamagata Univ.*)

We examined a response of the motility of actin filaments on heavy meromyosins (HMM) to an electric field applied between platinum electrodes in a motility assay system (25 mM KCl, 25 mM imidazole-HCl (pH 7.4), 4 mM $MgCl_2$, 1 mM ATP). When electrodes were set at 0.5 mm of gap size to which DC voltage was applied at 7 V, the velocity of actin filaments gradually decreased with the increase in time up to 12 s and they eventually stalled. Once the electric field was turned off, sliding velocity gradually increased again. Applying DC voltage for a further period induced the irreversible dissociation of actin filaments from HMM-coated surface. A higher voltage was necessary for the occurrence of these phenomena as the concentrations of KCl was decreased from 25 mM to 0 mM.

2Pos105 新規フォトクロミック阻害剤を用いたキネシン Eg5 の光可逆的制御

Photo-reversible control of mitotic kinesin Eg5 using a novel photochromic inhibitor

Ryoma Yamamoto, Kei Sadakane, Yuhki Tamura, Kentaro Saito, Shinsaku Maruta (*Soka University Graduate School of Engineering Division of Bioinformatics*)

Eg5 is one of the mitotic kinesin that maintain spindle formation and separate spindle. Inhibition of Eg5 results in and inducing cytostasis and apoptosis. It is well known that Eg5 has some specific potent inhibitors, e.g. STLC, Monastrol and Ispinesib. In this study, we designed and synthesized a novel photochromic inhibitor of Eg5, Maleimide Spiropyran-Cysteine (MASP-Cys) in order to control Eg5 photo reversibly. MASP-Cys exhibited reversible photo isomerization between spiro and merocyanine isomers upon visible light and ultraviolet light irradiations. MASP-Cys inhibited ATPase activity of Eg5 potently at less than 1μM of IC50. Merocyanine isomer of the inhibitor exhibited stronger inhibition than sprio isoform.

2Pos106 新規イネキネシン E11 に関する速度論的研究

Kinetic study on the novel rice plant kinesin E11

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In this study, we characterized biochemical and kinetic properties of another rice plant specific kinesin E11 that belongs to the plant specific At1 subfamily in kinesin-7 family. The fluorescent ATP analogue, Mant-ATP was employed for the kinetic characterization. We have successfully observed significant FRET between Mant-ATP and intrinsic tryptophan (Trp23) residue in E11. The kinetic parameters were analyzed by monitoring the FRET using stopped flow apparatus. The binding rate and dissociation rate were measured, and compared with other rice kinesin and conventional kinesin. The results revealed that the initial binding of ATP to E11 and release of ADP are faster than those of other rice plant specific kinesin.

2Pos107 速度改変型キメラミオシン XI の発現により明らかになってきた原形質流動の機能と制御

Function and regulation of cytoplasmic streaming on plant development

Motoki Tominaga[1], Kohji Ito[2], Takeshi Haraguchi[2], Etsuo Yokota[3], Akihiko Nakano[4,5] ([1]*Fac. Educ. Integrated Arts and Sci., Waseda Univ.*, [2]*Grad. Sch. Sci., Univ. Chiba*, [3]*Grad. Sch. Sci., Univ. Hyogo*, [4]*Grad. Sch. Sci., Univ. Tokyo*, [5]*RAP, RIKEN*)

Cytoplasmic streaming is active intracellular movement generated by organelle-associated plant-specific class XI myosin moving along the actin cytoskeleton in plant cells. However, its fundamental function in plants has remained unknown since its discovery more than 200 years ago.
To solve the query, we generated high- and low-speed chimeric myosin XI by replacing the motor domains of Arabidopsis myosin XI with those of the fastest Chara myosin XI and slower Human myosin Vb, respectively. Surprisingly, the plant sizes of the transgenic Arabidopsis expressing high- and low-speed myosin XI were larger and smaller than that of the wild-type plant, respectively. These results suggest that cytoplasmic streaming is a key determinant of plant size.

2Pos108 *Mycoplasma pneumoniae*・P1 adhesin の構造と機能

Structure and function of P1 adhesin of *Mycoplasma pneumoniae*

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Mycoplasma pneumoniae, a human pathogen glides with a unique mechanism. It forms a membrane protrusion, the attachment-gliding organelle at a pole and repeatedly catches and releases sialic acids on host cell surfaces. Here, we focus on the leg protein, P1 adhesin, which is a 170-kDa protein composed of three domeins and a transmembrane segment. We expressed and isolated this protein as a recombinant from Escherichia coli. The beads coated by this protein showed binding ability to sialic acids. Electron microscopy and image averaging showed that the molecule was shaped like headphone and about 10 nm in diameter. Now, we are analyzing a new construct fused with Strep-tag to achieve detailed analyses of binding ability and better alignments of electron microscopic images.

2Pos109 F1-ATPase の P-loop 変異体 TF1（βG158A）とリン酸解離の関係

The relationship between F1-ATPase P-loop mutant TF1(βG158A) and Pi release

Hiroka Narita[1], Hitoshi Hoshina[1], Hikaru Yoshida[1], Ayumi Ito[1], Jotaro Ito[1,3], Yohei Nakayama[1], Shoichi Toyabe[2], Hiroshi Ueno[1,3], Eiro Muneyuki[1] ([1]*Dept. of Physics. Chuo Univ.*, [2]*Faculty of Physics. Tohoku Univ.*, [3]*School of Engineering, The University of Tokyo*)

There are two schemes for the F1-ATPase motor. They differ in the order of Pi release after ATP hydrolysis. In 2010 Watanabe et al reported that F1 released Pi after ADP (Nat Chem Biol.2010 Nov;6(11)794-5). On the other hand, Shimo-Kon reported that Pi release occurred before ADP (Biophysical.J.98 1-10(2010)). In the present study, we tried to discuss this issue by examining a mutant F1-motor: a P-loop mutant in which hydrolysis of ATP and Pi release seem to be retarded. By single molecule rotation experiment using gold colloid, this mutant, TF1 (βG158A), exhibited a longer pause at catalytic dwell. Furthermore, analysis of the catalytic dwell of a chimeric F1 mutant including one mutant β or two mutant β suggested Pi release occurred before ADP release.

2Pos110 分裂期 kinesin-6 の運動特性に関する研究

A study on motility of mitotic kinesin-6

Yohei Maruyama[1], Akihiko Sato[1], Tim Davis[2], Toshihisa Osaki[3], Shin Yamaguchi[1], Shoji Takeuchi[3], Masanori Mishima[2], Junichiro Yajima[1] ([1]*Dept. Life Sci., Grad. Sch. of Arts and Sci., Univ. of Tokyo*, [2]*CMCB at Warwick Med. Sch., Univ. of Warwick*, [3]*Inst. of Ind. Sci., Univ. of Tokyo*)

Centralspindlin, kinesin-6 /CYK-4 complex, is essential for assembly of the central spindle. In this work, using 3D tracking microscopy, three dimensional movements of both kinesin-6 and centralspindlin along a suspended microtubule were quantified. We found that both kinesin-6 and centralspindlin displayed a left-handed spiraling motion around the microtubule. The rotational velocity of centralspindlin was slower than that of kinesin-6. There is no decrease in kcat value of centralspindlin in the ATPase activity assay, indicating that the reduction in the velocity was not due to a change of ATPase activity of kinesin-6 and that CYK-4 bound to a kinesin neck may cause the working distance of kinesin-6 to decrease by a change of the kiensin neck conformation.

2Pos111 ヒト細胞質ダイニン 1 分子のパワーストローク運動と反応機構

The power stroke mechanism of human cytoplasmic dynein revealed by optical tweezers

Yoshimi Kinoshita[1], Taketoshi Kambara[2], Kaori Nishikawa[1], Motoshi Kaya[1], Hideo Higuchi[1] ([1]*Graduate School of Science, The University of Tokyo*, [2]*RIKEN QBiC*)

Cytoplasmic dynein is a motor protein moving along microtubules toward the minus-end, and plays an important role in cellular processes. Dynein's conformational change, called power stroke, is assumed to generate driving forces moving along the microtubule. However, it has not been clarified the mechanism of how the power stroke contributes to individual steps. Therefore, we measured the distance driven by the power stroke and the dwell time that single-headed human cytoplasmic dynein binds to a microtubule with optical tweezers. We evaluated the dependence of nucleotide on these values, and discuss the mechanochemical cycle of dynein with a focus on the power stroke mechanism.

2Pos112 DNA オリガミバネを用いた負荷存在下でのキネシンの運動の蛍光一分子観察

Single-molecule fluorescent observations of kinesin-1 moving under a load from DNA origami spring

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Kinesin-1 is a motor protein that moves along microtubule to transport cargoes inside the cells. Single molecule motilities of kinesin under load have been extensively studied using optical trapping assays, although it has been difficult to simultaneously observe single molecule fluorescent. To elucidate the mechanism of force production, it is important to monitor conformational changes using fluorescent-based methods under external load. Here we used coil-shaped DNA origami spring and immobilized one end to microtubule via immotile kinesin and attached Qdot-labeled kinesin to the other end. We observed saw tooth-like traces of the dye similar to the optical trapping assays; kinesin moves up to 1 μm and then stalls until it detaches from the mictotubule.

2Pos113 長円形 PDMS チャンバー内における微小管の動的不安定性

Dynamic instability of microtubules observed in oval PDMS microchambers

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Local effects such as steric hindrance and chemical gradient are the keys to understanding of cells. Our goal is to mimic enhancement of local effects and simultaneously estimate localization of components for dynamic instability of microtubules (MT) *in vitro*. Dynamics of MT were observed in oval PDMS microchambers (1x1x15 μm) under the TIRF microscope. GTP-bound tubulin dimers gradually polymerized onto GMP-CPP-stabilized biotin-labeled MT seeds which are initially attached to the PEG-biotin-coated glass. When encapsulated in oval chambers, dynamics of MT seem to be affected by rapid changes in concentration of components inside chambers. Moreover, dead-end effects seem to be present. These features are compared with those in conventional wide-open flow cells.

2Pos114 高分解能ステップ計測で分かってきた細菌べん毛モーターのサーマルラチェット機構

若手招待講演

Thermal ratchet mechanism of the bacterial flagellar motor emerged by high-resolution nanophotometry

Shuichi Nakamura[1,2], Yusuke V. Morimoto[2,3], Nobunori Kami-ike[2], Yoshiyuki Sowa[4], Tohru Minamino[2], Keiichi Namba[2] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*QBiC, RIKEN*, [4]*Dept. Frontier Biosci., Hosei Univ.*)

The bacterial flagellar motor is a rotary nanomachine driven by proton-motive force. Interaction between a rotor and a stator is responsible for torque generation, but the rotation mechanism remains unclear. Here we observed stepping rotations of flagellar motor rotating at ~300 Hz by extremely high-resolution nanophotometry capable of determining the position of a 100 nm gold nanoparticle at 1 nm resolution at a frame rate of 390 kHz. Step speeds were widely distributed, reaching ~4,000 Hz. The step-speed distribution of the wild-type motor appeared to be Boltzmann distribution, which was similar to that of the stator-less mutant motor showing diffusive rotation. These data suggest that the flagellar motor is a thermal ratchet driven by the Brownian motion.

2Pos115 好アルカリ性 *Bacillus* 属細菌が持つ Na^+駆動型べん毛モーター固定子 MotPS の中性環境でのイオン透過機構の解明

Analysis of the reduced motility mechanism of the Na^+-driven flagellar motor stator MotPS in alkaline *Bacillus* at low pH

Yuka Takahashi[1], Yukina Noguchi[2], Masahiro Ito[1,2] ([1]*Bio-Nano., Toyo Univ.*, [2]*Grad. Sch. Life Sciences, Toyo Univ.*)

The flagellar motor is generally energized by either a H^+ or Na^+ motive force. Alkaliphilic *Bacillus pseudofirmus* OF4 has a MotPS as a stator and the flagellum is driven by a Na^+-motive force. Previous studies showed that the motility of OF4 is dependent on the Na^+ concentration at pH above 8, but little motility was observed at neutral pH even in the presence of high Na^+ concentrations. Our current studies suggested that MotP-N186 from OF4 may be critical for regulation of Na^+ influx at low pH. Therefore, we constructed a mutation at the same positon of MotP-N186 in other *Bacillus species*. The data suggest that this site is critical for Na^+ influx at low pH.

2Pos116 人工繊毛モーターの創製ー微小管の波打ち運動発現条件ー

Development of self-propelled artificial cilia constructed from biomolecular motors

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Biomolecular motor system microtubule-kinesin can transform chemical energy into mechanical work. Since kinesin has a higher efficiency compared to man-made motors, considerable efforts have been devoted to develop artificial devices using microtubule-kinesin system. However, most of the efforts were concerned about the generation of the gliding motion of microtubules on a two dimensional surface. In nature, ciliated protists such as paramecium can move freely underwater by utilizing oscillatory beating of cilia as motive force. Here, I aim at developing artificial cilia mimicking the swimming of the paramecium. To utilize the beating of microtubules as motive force, we first optimize conditions to generate beating of mictotubules.

2Pos117 超好熱菌 *Aquifex aeolicus* の固定子タンパク質 MotA のナノディスク再構成とその機能解析

Functional reconstitution of stator protein MotA from *Aquifex aeolicus* into Nanodisc

Mizuki Gohara[1], Norihiro Takekawa[1], Naoya Terahara[2], Takayuki Kato[2], Keiichi Namba[2,3], Yasuhiro Onoue[1], Michio Homma[1] ([1]*Divi. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*, [2]*Grad. Sch., Frontier Biosci., Osaka Univ.*, [3]*Riken QBiC*)

The stator complex in the bacterial flagellar motor forms a specific ion-conducting pathway. The ion flux of the stator couples to torque generation through its interaction with the rotor. The stator of the flagellar motor consists of 4 MotA (PomA) and 2 MotB (PomB) molecules. Surprisingly, our previous overexpression and purification results suggested that MotA (PomA) can form a stable oligomer complex without MotB (PomB). In this study, by using MotA from *Aquifex aeolicus* and PomA from *Vibrio alginolyticus*, we demonstrate successful reconstitution of MotA (PomA) into Nanodisc. Currently we are carrying out structural and biochemical experiments on these functionally reconstituted stators composed of MotA (PomA) only.

2Pos118 生細胞内における微小管へのキネシン結合速度定数の直接計測

Direct measurement of the binding rate constant of kinesin to microtubules in living cells

Taketoshi Kambara, Yasushi Okada (*Riken, Quantitative Biology Center*)

It has been established that conventional kinesin, KIF5, selectively moves along a specific subset of microtubules in living cells. However, the mechanism of this selective binding is still controversial. To understand the mechanism of the selective binding, it would be important to examine whether kinesin binding to specific subsets of microtubules is enhanced, inhibited or both. Here, we directly measured the binding rate constant (*kon*) of kinesin to microtubules in living cells using single molecule fluorescence microscopy. To our surprise, *kon* of KIF5 to microtubule in vivo was nearly 3~5 times higher than that in vitro, suggesting that mechanisms exist in the cell to recruit KIF5 specifically to some subset of microtubules by accelerating the binding reaction.

2Pos119 Measurment of the transition between docking and undocking of kinesin neck linkers

Yuichi Kondo, Hideo Higuchi (*Grad. Sci. Phys., Univ. Tokyo*)

Kinesin-1 is a double-headed molecular motor that moves processively along a microtubule toward the plus end. The neck linker docking model that kinesin motility is biased forward by the neck linker docking to the motor head is widely accepted. However it is unclear how the neck linker docking contributes to the processive motion and the force generation. To address the contribution quantitatively, we observed the transition between docking and undocking states and determined the size of conformational change by single-molecule measurement using optical tweezers. We show that the docking and undocking events are load dependent and discuss the mechanism of the kinesin movement.

2Pos120 FRAP による、成長円錐のラメリポディア領域におけるファシンの動態解析

Dynamics of fascin analyezed by FRAP, in the lamellipodial region of the growth cone

Minami Tanaka[1,2], Ryoki Ishikawa[3], Kaoru Katoh[2] ([1]*Grad. Sch. Life & Env. Sci., Univ. Tsukuba*, [2]*Bio Mes Res. Inst., AIST*, [3]*Gunma Pref.Coll. Health Sci*)

Growth cones are enlargement of cytoplasm which appear at the tip of growing neurites. The growth cone movement plays an important roles in path finding and neuronal navigation. Sensory mechanism for the target recognition is present at the tip of filopodia. The movement of growth cone is controlled by actin dynamics.
Filopodial formation is controlled by fascin, an actin bundling protein, The activity of fascin depends on phosphorylation.
We, therefore, performed FRAP experiments on GFP labeled actin, fascin and mutant fascins. We found a molecular mechanism of phosphorylation-dependent binding of fascin. Detailed data and images will be shown in our presentation.

2Pos121 Dynamical Patterns and Physical Environments in Vibrio alginolyticus Swarm Plate

Tzu-Jung Hsu (*National Central University*)

The aim of this work is investigating dynamical patterns and the physical environments of bacterial swarming on the agar plates. When Vibrio alginolyticus grow on the agar plates, it will sense the surface and transform into swarming cells expressing high density lateral flagella and elongating without division. We measure the physical properties such as effective viscosity of the agar plates and colony surface profile to explain the physical original of this transformation.
The collective motion of these elongated cells shows dynamical patterns including waving dynamics and jamming process. These two dynamical patterns can be explained by the self-propelled particle theory. We will demonstrate our latest observation and explanation.

2Pos122 *Spiroplasma eriocheiris* において観察されたキンクの伝搬によって生じるらせん型スクリューの向きと回転速度の変化
Change in handedness and rotational speed of helical screw of *Spiroplasma eriocheiris* driven by kink propagation

Tatsuro Itou[1], Yoshiaki Kinosita[1], Daisuke Nakane[1], Hirofumi Wada[2], Wen Wang[3], Takayuki Nishizaka[1] ([1]*Department of Physics, Gakushuin Univ.*, [2]*Department of Physics, Ritsumeikan Univ.*, [3]*College of Life Science, Nanjing Normal Univ.*)

Spiroplasma are helical shaped bacteria. They swim in one direction roughly parallel to a long axis of the cell body, and there appear several kink-like nods propagating from the front end to rear end. To unravel the mystery of the kink, we determined the handedness of the screw and recorded the swimming motility of *Spiroplasma eriocheiris* under an optical microscope by a high-speed camera. The handedness was inverse before and after the kink, and the screw did really rotate to reach 17 Hz at maximum, presumably driven by the kink propagation. We estimated, under the viscosity with the range of 10-1000 mPa s, various parameters including the speed of cell swimming, screw rotation and kink propagation; the pitch of the screw; and the distance between multiple kinks.

2Pos123 クライオ電子線トモグラフィ法を用いたフィロポディア内アクチンフィラメント束化メカニズム解明
Revealing the bundling mechanisms of actin filament in filopodia with cryo-electron tomography

Shinji Aramaki[1], Kouta Mayanagi[2], Kazuhiro Aoyama[3,4], Takuo Yasunaga[1] ([1]*Dept. of Bioscience and Bioinformatics, Kyushu Inst. of Tech.*, [2]*Medical Inst. of Bioregulation Kyushu Univ.*, [3]*FEI Company*, [4]*Research Center for Ultra-High Voltage EM, Osaka Univ.*)

Filopodia are important cell membrane protrusion structure for many cellular functions, and they have very characteristic structure which thickness is around 200-400 nm and actin filaments rich structures. Despite of importance of them, formational and functional mechanisms of them are still unknown. In order to reveal the mechanisms of them we adopted the cryo-electron tomography (cryo-ET) technique. These techniques provided us the three-dimensional models of filopodia without any artifacts from chemical fixation at molecular resolution. The three-dimensional data of filopodia told us actin filaments bundling properties in filopodia. These novel knowledges will help us to understand formational and functional mechanisms of filopodia.

2Pos124 マウス網膜細胞光シグナル伝達の生体分子混み合いを考慮した数理モデル
Rate equation models of phototransduction influenced by molecular crowding into membranous disks of mouse rod cell

Rei Takamoto[1], Hiraku Nishimori[1,2], Akinori Awazu[1,2] ([1]*Department of Mathematical and Life Science, Hiroshima University*, [2]*Research Center for Mathematics on Chromatin Live Dynamics, Hiroshima University*)

Mouse rod phototransduction process is investigated through rate equations models of biochemical reactions of several molecules that contributie to the signaling process into the membranous disks in outer segments of rod cell. By considering the molecular crowding effects of rhodopsin as expected by experiments that may hinder the diffusion and reactions of molecules on disk membrane, our models can reproduce the following phenomena observed in experiments; I) The activations and relaxation of this signaling pathway of WT mouse rod cell are slower than those of mutant cell involving a half of rhodopsin on membrane. II) Relaxation time after the photo-activation of this signaling pathway exhibits nonlinear dependencies of input light-strength.

2Pos125 ケラトサイトのかたち形成におけるストレスファイバーの役割
Stress fibers contribute to the shape determination in fish keratocytes

Takako Nakata[1], Chika Okimura[1], Takahumi Mizuno[2], **Yoshiaki Iwadate**[1] ([1]*Fac. Sci., Yamaguchi Univ.*, [2]*Biomed. Res. Inst., AIST*)

Crawling cells have characteristic shapes dependent on their cell-types. How they determine their shape is interesting issue. Fish keratocytes maintain nearly constant fan-shape during their crawling locomotion. We compared the shape and related molecular mechanisms in keratocytes from different fish species to reveal what mechanism is a key factor for the cell shape determination. Wide keratocytes from cichlid exerted large traction forces at the rear ends and showed spatially loose gradient of actin retrograde flow rate. Whereas, round keratocytes from black tetra exerted small traction forces there and showed steep gradient of it. The power source, stress fibers might play an important role for maintaining cell shapes by regulating actin retrograde flow rate.

2Pos126 アクチンフィラメントとアクチン結合タンパク質間の協同的結合の経時的観察
Longitudinal observation of cooperative binding between actin-binding proteins and actin filament

Rika Hirakawa[1], Atsuki Yoshino[1], Keitaro Shibata[2], Hiroaki Ueno[1], Taiga Imai[1], Taro Q.P. Uyeda[2], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Sustain. Environ. Eng., Muroran Inst.*, [2]*Biomedical Res. Inst., AIST.*)

Cooperative binding between actin-binding proteins and actin filament is attributed to that the binding of the proteins to the filaments induces cooperative conformational changes of neighboring actin subunits. The purpose of this study is to analyze the propagation of the cooperative conformation change that is evoked by binding of the actin-binding proteins. In this study, we continuously observed the binding of fluorescent protein-fused actin-binding proteins, which were myosin, cofilin, fimbrin, talin, and drebrin, to immobilized actin filament on lipid membrane using fluorescence microscopy. The results suggested that the cooperative binding of myosin, cofilin, and fimbrin was successive as a domino effect.

2Pos127 **Photo-regulation of small G protein RhoA using photochromic molecules**

Masahiro Kuboyama[1], Kaori Masuhara[2], Shinsaku Maruta[2] ([1]*SOKA University Department of Bioinfomatics*, [2]*SOKA University Graduate School of Engeneering*)

Aim of this study is to control the interaction of small G protein RhoA with its downstream effector Rho-associate kinase2 (Rock2) using photochromic molecules photo-reversibly. Previously, we have shown that GTPase activity of K-Ras which shares common molecular mechanism with RhoA is photo-reversibly controlled by the isomerization of photochromic molecules, 4-phenylazophenyl maleimide (PAM) incorporated into the K-Ras functional sites. In this study, we prepared RhoA mutants that have a single cysteine residue at the functional site using Bac-to-Bac expression system and modified with photochromic azobenzene and spiropyran derivatives. Photo-regulation of the interaction of the modified RhoA with Rock2 was monitored by the method of pull down assay.

2Pos128 **局在した細胞間接着が生み出す細胞運動パターン**
Localized intercellular adhesion inducing moving cell pattern

Katsuyoshi Matsushita (*CMC, Osaka Univ.*)

Dictyostelium Discoideum (Dicty) is a good model system for the study of the cell motion. In the motion, the intercellular adhesion promotes the collective behavior of cells. In the adhesion, an adhesion protein DdCAD1 plays an important role in an early stage of the fruiting body formation of Dicty. The protein is known to localize on pseudopods of Dicty. The effect of the localization of the protein on the collective cell migration is not well known. We explore effects of the localization on the cell motion by using a numeical model. We find that the localization induces a pattern of moving cells. We report the result.

2Pos129 **Structural characterization of FliP, a component of flagellar type III protein export apparatus**

Takuma Fukumura[1], Fumiaki Makino[1], Takayuki Kato[1], Katsumi Imada[2], Keiichi Namba[1], Tohru Minamino[1] ([1]*Grad. Sch, Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Sci. Osaka Univ*)

Many component proteins of the bacterial flagellum are transported by a specific export apparatus from the cytoplasm to the growing distal end of the flagellum for self-assembly. The export apparatus consists of a water-soluble ATPase complex and a proton-driven export gate made of six membrane proteins (FlhA/B, FliO/P/Q/R). Recent genetic and biochemical analyses have shown that FliP is thought to assemble at an earliest stage of export gate formation. We overexpressed and purified FliP of Salmonella. EM observation revealed a ring-shaped hexamer with a central pore of 2 nm in diameter. The crystal structure of the periplasmic domain of Thermotoga maritima FliP (Tm-FliPP) determined at 2.4 Å resolution suggests that FliP dimer is a functional unit.

2Pos130 **柔軟性に富んだ微小管内 GTPγS-チューブリン分子**
X-ray fiber diffraction analysis revealed a highly flexible state of GTPγS-tubulin in the microtubules

Shinji Kamimura[1], Yosuke Fujita[1], Yuuko Wada[1,2], Tomohiro Shima[3], Yasushi Okada[4], Hiroyuki Iwamoto[5] ([1]*Dept. Biol. Sci., Chuo Univ.*, [2]*Inst. Global Leader.*, [3]*Dept. Biol. Sci., Grad. Sch. Science, Uni. Tokyo*, [4]*QBiC, Riken*, [5]*JASRI, SPring-8*)

Our major question is how tubulin states are related to the stability of microtubules. To address the question, we used our novel technique for the quick shear-flow alignment of biological filaments, which enabled us to acquire fine X-ray fiber diffraction signals from native microtubules in a few seconds. We found that microtubules could be classified into three major groups with distinct axial periodicities of tubulin, which varied depending on the temperature of solution, the state of GTP-hydrolysis and the contents of microtubule stabilizers. It was also revealed that the GTPγS-tubulin showed the widest variation in the longitudinal tubulin periodicity in situ, suggesting a highly flexible state of GTP-tubulin in the microtubules.

2Pos131 **細胞ネットワーク計測のための拡張可能なモジュール型多電極細胞外電位計測システムの開発**
Development of flexible expandable on-chip multi electrode array system for cell-network measurement

Fumimasa Nomura, Akihiro Hattori, Kenji Matsuura, Hiromi Kurotobi, Masao Odaka, Hyonchol Kim, Hideyuki Terazono, Kenji Yasuda (*Institute of Biomaterials and Bioengineering, Tokyo Medical and Dental University*)

To measure the invisible electrophysiological properties among cells in their networks non-destructively, we have newly developed a flexibly expandable multi electrode array (MEA) system. This system is composed of a multiple of four channel expandable units to meet to the desired number of measurement/stimulation channels for the experiments. We constructed the line-up cardiomyocytes network on the MEA chip using agarose-gel microstructures, and obtained the conduction velocities and the action potential duration from four electrodes and compared. The results indicated the fluctuation of electrophysiological propagation can predict the action potential duration as a simple and new measurement method of electrophysiological analysis.

2Pos132 **細胞性粘菌の集団運動における細胞極性の動態**
The dynamics of cell polarity during collective migration of *Dictyostelium* cells

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From fibroblasts to neutrophils, cells are known to frequently reorient by extending protrusions in random orientation. How such spontaneous dynamics are suppressed or utilized in collective cell migration is unclear. We analyzed the chain migration of *Dictyostelium* cells by live cell imaging. Here, we show that there is a unique pattern of F-actin localization characterized by its enrichment at the periphery of the cell-cell contact region. Knockout of TgrB1/C1 resulted in the loss of persistent F-actin pattern at the contact site and decreased occurrence of the chain migration. These results suggest that TgrB1/C1 adhesion promotes continuous actin polymerization, which enables to extend protrusions cooperating with the front cell migration.

2Pos133 1細胞分泌実時間イメージングの並列測定プラットフォーム
Parallel measurement platform for real-time single-cell secretion imaging

Kaede Miyata[1], Yoshitaka Shirasaki[1,2], Nobutake Suzuki[1,2], Mai Yamagishi[1,2], Sotaro Uemura[1] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*IMS, RIKEN*)

In bulk measurements, a large population of cells shows particular secretory response depending on the dose, kinds or timing of stimulation. On the other hand, it is known that single cells show heterogeneous secretory responses for an input even if they are cloned populations. The question is how the specificity of ensemble response is achieved in spite of great individuality. To address this issue, characters and variations of responses from individual cells are to be compared between different stimulation under the same cell conditions. In this meeting, we will talk about the development of the parallel measurement platform for real-time single-cell secretion imaging with the multi-reservoir integrated nano litter-well array chip.

2Pos134 三次元で見たATP依存的な繊毛運動のCa^{2+}による制御
Regulation of ATP-dependent ciliary motility by Ca^{2+} observed in three dimensions

Toshihito Iwase[1], Masaaki Suegara[1], Rinako Nakayama[2,5], Takanobu A Katoh[3], Mitsutoshi Setou[4], Takayuki Nishizaka[3], Koji Ikegami[4], Tomoko Masaike[1,2,5] ([1]*Dept. Appl. Biol. Sci., Tokyo Univ. of Sci.*, [2]*Res. Inst. for Sci. and Tech., Tokyo Univ. of Sci.*, [3]*Dept. Phys., Gakushuin Univ.*, [4]*Dept. Cell Biol. and Anat., Hamamatsu Univ. Sch. Med.*, [5]*PRESTO, JST*)

Exclusion of harmful materials from trachea is supported by the increase in frequency of ciliary beating. To investigate regulation of ATP-dependent ciliary motility by Ca^{2+}, we observed beating of individual demembranated cilium from trachea of mice under the 3D tracking microscope with a fluorescent bead attached to the tip as a probe. Increased beating amplitude and velocity but unchanged frequency (about 6 Hz) were observed at higher Ca^{2+} concentrations in the range of 10-1000 nM. On the contrary, beating frequency was clearly dependent on ATP concentration. Our results explain previous reports that beating frequency was affected by Ca^{2+}. We hypothesize that the increase in concentrations of ATP triggered by Ca^{2+} is the direct cause of high beating frequency.

2Pos135 サルモネラべん毛モーターにおけるMotA Met-206の役割
Role of MotA Met-206 in *Salmonella* flagellar motor

Kodai Oono[1], Shuichi Nakamura[1], Fumio Hayashi[2], Kenji Oosawa[3], Seishi Kudo[1] ([1]*Grad. Sch. Eng., Tohoku Univ.*, [2]*Instrumental Analysis Center., Gunma Univ.*, [3]*Div. Mol. Sci., Fac. Sci. and Tech, Gunma Univ.*)

The bacterial flagellar motor converts proton influx through a stator into rotation. The stator consists of MotA and MotB. The interaction between MotA and a rotor protein FliG is involved in torque generation, but the rotation mechanism is unclear. Here we performed the rotation analysis of a *Salmonella* MotA mutant motor with a point mutation M206I. The torque-speed curve showed that M206I motor can generate torque at the wild-type level at high load but its rotation rate at the low load was much slower than the wild-type motor. The rotation rate of M206I motor increased with decrease of external pH. These results suggest that the MotA Met-206 plays a key role in protonation of stators and that the proton translocation is not directly coupled with torque generation.

2Pos136 周期的伸展刺激に対する心筋細胞の形態や配向性の変化
Morphological and orientation change of cardiomyocytes to cyclic stretch stimulation

Chiho Nihei, Tomoyuki Kaneko (*LaRC, Dept. Frontier Biosci., Hosei Univ.*)

Cardiomyocytes are subjected to mechanical stress constantly in vivo as hearts continually contract and relax. In this examination, we applied mechanical stress to cardiomyocytes derived from chicken embryo (E13) by cyclic stretch stimulation. Agarose on polydimethylsiloxane (PDMS) stretch chamber was microfabricated by infrared laser, then were arranged cardiomyocytes on the agarose micro chamber. After cardiomiocytes adhered on the chamber, cyclic stretch stimulation was applied to combination of two conditions of cycle and magnitude of stretching.Thereafter, we observed morphological and orientation change of cardiomyocytes.

2Pos137 原子間力顕微鏡による肝細胞共培養系の力学特性解析
Mechanical analysis of hepatocyte coculture system using atomic force microscopy

Ryosuke Tanaka[1], Yoshikatsu Akiyama[2], Jun Kobayashi[2], Masayuki Yamato[2], Takaharu Okajima[1] ([1]*Grad. Sch. Info. Tech., Hokkaido Univ*, [2]*Inst. Adv.BioMed. Eng. Sci., Tokyo Women's Medical Univ*)

Hepatocyte-specific functions such as albumin and urea synthesis are enhanced, being co-cultured with endothelial and fibroblast cells [1] and are also affected by adhered cell area [2]. These results suggest that hepatic functions are correlated with the mechanical property of co-cultured cells, but the correlation is not known. Here, we investigate the elastic modulus E of co-cultured hepatocytes with endothelial or fibroblast cells by atomic force microscopy. We found that E of co-cultured hepatocytes significantly changed over a range in a few mm-scale, indicating that hepatic-functions are regulated by a long-range mechanical interaction between co-cultured cells. [1]Tsuda et al., Biochem Biophys Res Commun. 29 (2006) 937. [2] Singhvi et al, Science, 264(1994) 696.

2Pos138 アクチンストレスファイバー内ミオシンのダイナミクス
Dynamics of nonmuscle myosin molecules in actin stress fibers

Tsubasa S. Matsui, Tomoya Ikeda, Shinji Deguchi (*Dept. Nanoparm. Sci., NITECH*)

In nonmuscle cells, contractile forces play impotant roles in broad aspects of cell functions. The forces are mainly generated in actin stress fibers composed of nonmuscle myosins and actin filaments, and the force level is regulated by the myosin ATPase activity. The ATPase activity is thought to be modulated by two cues: the phosphorylation state of myosin regulatory light chain (MLC) and load-sensitive myosin ATPase cycle. Here, to reveal the relationship between the biochemical and physical cues, we investigate the turnover rates and fractional recovery of myosin molecules associated with phosphomimic and nonphosphorylatable MLC mutants (collaborative work with Dr. Masayuki Takahashi, Hokkaido Univ.).

2Pos139 海洋性ビブリオ菌べん毛モーター FliL 蛋白質の生物物理性質と機能解析

Analysis of function and biophysical properties of marine *Vibrio* FliL in the flagellar motor

Ananthanarayanan Kumar, **Yuuki Nishino**, Shiwei Zhu, Mayuko Sakuma, Seiji Kojima, Michio Homma (*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*)

Many bacteria can swim using flagella. Rotation of the flagella motor is driven by the interaction between rotor and stator. There is one transmembrane flagellar component, named FliL. FliL is an 18kDa, single transmembrane component and is involved in the torque generation. We investigated the role of FliL in the sodium-driven polar flagellar motor of *Vibrio alginolyticus*. In *V. alginolyticus*, FliL localizes at the one pole and its localization depends on that of stator. We found that FliL affected function of flagellar motor in which MotB-PomB chimera or plug deleted PomB mutants were expressed. We also purified soluble periplasmic region of FliL and succeeded in its crystallization. We will discuss the function and biophysical properties of FliL.

2Pos140 ビブリオ菌べん毛モーター MS リング構成因子 FliF の精製タンパク質会合状態

Oligomeric states of purified FliF, a MS-ring component of Vibrio flagellar motor

Erika Yamaguchi, Seiji Kojima, Michio Homma (*Div. Bio. Sci., Grad. Sch. Sci., Nagoya Univ.*)

The MS-ring is a part of the flagellar basal structure embedded in the inner membrane and composed of membrane protein FliF. It has two transmembrane segments and a large periplasmic region. When *Vibrio alginolyticus* FliF is overproduced in *E. coli*, FliF is equally detected from soluble and membrane fractions. Size exclusion chromatography (SEC) showed that FliF from both fractions behave as oligomer. We purified FliF at several conditions and evaluated oligomeric states of FliF using Dynamic Light Scattering (DLS). We found that purified FliF showed the smaller radius as judged by SEC and DLS analyses when solubilized using Decylmaltoside or at alkaline condition. We hope further characterization of FliF reveal the unique feature for the MS-ring assembly.

2Pos141 細胞周期での細胞の大きさや形から得られる新たな情報

New obvious information obtained from the cell size and shape during mitosis cycle

Rina Nagai[1,2], Keisuke Ohta[1,3], Takako Ichinose M.[1,2], Hikari Mori[1,2], **Atsuko Iwane H.**[1,2] ([1]*Cell Field Struc., QBiC, Riken*, [2]*Spec. Res. Promot. Group, Grad. Sch. Fronti., Biosci., Osaka Univ.*, [3]*Anatomy, Med., Kurume Univ.*)

EM offers exceptional resolution of extremely small biological specimens, providing images of the minutest of organelles and molecules responsible for fundamental cellular phenomena. FIB-SEM will provide us information of the specimen which will complement the 3D-structural information. We selected C. merolae cell, a primitive red algae, as a model organism for cell mitotic cycle. Although the size of the cell is 2-5 micron, the body shape is not well understood yet. In this meeting in addition to analyze the interaction between individual several organelles during mitosis cycle, we aim to reveal individual morphology details and compute the volume occupancy of each organelle. We will discuss you about the relationship between cell body mass and energy efficiency, also.

2Pos142 Bombinin H2 および H4 の細菌模倣膜に対する相互作用の解析

Analysis of interaction of Bombinin H2 and H4 with the mimetic bacterial membrane

Shiho Kaneda, Yuki Kitahashi, Akira Naito, Izuru Kawamura (*Graduate School of Engineering, Yokohama National University*)

Bombinin H2 and its diasteromer Bombinin H4, which are difference in the second amino acid with L-Ile in H2 and D-allo-Ile in H4, are cationic antimicrobial peptides from frog skin of Bombina variegata. We researched interactions of peptides with the mimetic membrane containing cardiolipin lipids like Staphylococcus aureus. In mass experiments using cross-linker BS3 at 40°C, we observed peptide peaks of monomer and dimmer for any case of H2, H4 and H2-H4 mixed sample. This result suggested that peptides interact each other in the membrane. In ^{31}P solidstateNMR measurements, NMR signals for any systems appeared at 0 ppm at 10°C. This results indicated that the mimetic membrane with peptide was disrupted. Therefore, the antimicrobial activity of peptides was induced.

2Pos143 FTIR-ATR プリズム上に作製したセラミド/ステロール混合膜への重水透過性

Heavy water permeation into the ceramide/sterol mixed film prepared on an FTIR-ATR prism

Kohei Oka, Satoru Kato (*Grad. Sch. Sci & Tech., Univ. Kwansei Gakuin*)

We examined heavy water permeation into the lipid film composed of ceramide and cholesterol analog to clarify the role of the cholesterol molecular architecture in the barrier function. The permeability was evaluated by a newly developed method. The ceramide/cholesterol analog mixed film was prepared by spraying the lipid solution onto an FTIR-ATR prism to form a homogeneous film. After a drop of heavy water was mounted onto the lipid film, the FTIR peak centered at 2500 kayser increased exponentially as the heavy water molecules reached to the vicinity of the prism surface. We estimated the permeability from the time constant for the exponential growth of the FTIR signal and will discuss its dependence on the molecular architectures of cholesterol analogs.

2Pos144 セラミドホスホエタノールアミンはマイクロメートルサイズのらせん構造をつくる

Microstructural Polymorphism of Ceramide Phosphoethanolamine

Takehiko Inaba, Motohide Murate, Yan-Fen Lee, Francoise Hullin-Matsuda, Peter Greimel, Toshihide Kobayashi (*RIKEN Lipid biology laboratory*)

Lipids spontaneously form bilayers and bilayers close the edge and form a lipid vesicle (liposome). In some case, lipids form the other structures depend on lipid species, solvent, and forming process. We focused on the structures made from ceramide phosphoethanolamine (CPE). CPE is the major sphingolipid in insect cell and the sphingomyelin analog which is used for glial cell. To reveal the microstructure of CPE, darkfield microscopy was used to overcome the detection limit of thin lipid structures without fluorescent labelling. We found the condition that helical structures were observed in the composition of PC and CPE mixture. This result suggest that a polymorphism derived from CPE might be used for insect cell membrane morphogenesis.

2Pos145 DNA 構造の自己組織化によるマイクロサイズカプセルの形成

Microsize capsule formed by self-assembly of DNA structures

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3D architectures built of biomolecules have been attracting much attention as a viable approach to create minute robots with sensing, diagnostics and directed motion. 3D DNA nanostructure is a powerful tool to produce molecular robots because of capability of designing intended structures with selective functionalization followed by programmed assembly of 2D DNA nanostructures. However, because a piece of 2D DNA structure has the size of only a few hundred of nanometers, microscale architectures have not been achieved so far. If we form a larger-size 3D DNA architecture, practical applications for various materials will be implemented. Herein, we present a microcapsule composed of 2D DNA nanostructures formed at the water-oil interface.

2Pos146 細胞膜へのぬれ性がゲル弾性へ及ぼす影響

How membrane wetting affects elasticity of biopolymer gels in model cells?

Atsushi Sakai, Miho Yanagisawa (*Dept. Appl. Phys.,Tokyo Univ. of Agri. & Tech.*)

Biomembrane and actin cytoskeleton undergo intimate interplay that is responsible for morphogenesis. Although the gel elasticity has been investigated, effects of membrane and inhomogeneous cytoplasm on the elasticity are unclear. To reveal such effects, we prepare a model cell with a cytoplasm in so-gel phase: PEG and gelatin droplet coated with a lipid layer. The interactions with the membrane vary wettability of the gel. In the case of a partial wetting, the gel shape is a hemisphere. We analyze the gel elasticity using micropipette aspiration. Consequently, the wetting side is much stiffer than the phase boundary side. It means that phase equilibrium of the polymer blend confined in model cells differs from that in bulk, which may lead to the elastic inhomogeneity.

2Pos147 ナノ粒子と質量分析装置を用いて特定タンパク質周辺の脂質を分析する方法の開発について

Development of a new method to detect lipids surrounding specific proteins using nanoparticles and mass spectrometry

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In biomembranes, there are spatial heterogeneity of lipid composition like lipid raft. These heterogeneities support many proteins' functions. Despite the importance of the heterogeneities, there are no direct method to detect endogenous lipids around specific protein because the size of these heterogeneities are very small. In this research, we are developing a new method that analyzes the lipids which surround the antigen proteins by using antibody-labeled nanoparticles and MALDI -TOF MS equipment. We applied this method to rhodopsin and CD59. As the results, we detected several kinds of fatty acids which composing phospholipids which surround the antigen proteins by using covalently linked gold nanoparticle, antibody and antigen complex.

2Pos148 固体基板が誘起する人工脂質膜の非対称性と異方性

Substrate-Induced Asymmetry and Anisotropy of Molecular Diffusion in Artificial Lipid Membranes

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The purpose of this study is addition of heterogeneous structure to substrate-supported lipid bilayers (SLB) by the substrates effects. We investigated the single molecule diffusion in SLBs on three single-crystal substrates: SiO2/Si, mica and sapphire. The diffusion of single dye-lipids in the SLBs was observed by diagonal illumination set-up. In the SLB on SiO2/Si, the single and isotropic diffusion component was obtained. That on mica showed double components, which indicate the decoupling of upper and lower leaflets in SLB. On sapphire, the anisotropic diffusion was obtained because the atomic steps acted as potential barrier. The quantitative evaluation of substrate effects on SLBs provides a new strategy for the creation of biomimetic membranes.

2Pos149 ヌクレオチドに誘起される巨大細胞膜小胞の形態変化

Nucleotides induce morphological change of giant plasma membrane vesicles

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Giant plasma membrane vesicles (GPMVs) are one of the cell membrane model which are separated from cultured cells directly. The GPMVs seem to maintain lipids and membrane proteins. So, we used this membrane model to study physical properties of lipids and membrane proteins *in vitro* .

In this study, we have tried to find reagents that induce morphological change of GPMVs to understand a molecular mechanism of cell membrane shape change. We found the interestingly phenomenon that nucleotide can induce morphological change of GPMVs from spherical shape to odd shape like stretched dumbbell. This change may be induced by insertion of the nucleotide molecules to the GPMVs. We will discuss how nucleotide drive morphological change of GPMVs.

2Pos150 単一 GUV 法を用いた細胞透過ペプチド・ポリアルギニンの脂質膜ベシクルへの侵入の研究

Investigation of the Entry of Cell-Penetrating Peptide, Polyarginine into a Vesicle of Lipid Membrane Using the Single GUV Method

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To elucidate the mechanism of the entry of polyarginine (Rn) into cells, we investigated its entry into a vesicle by using single giant unilamellar vesicle (GUV) method [1]. The interactions of carboxyfluorescein labeled, R9 (CF-R9) with single DOPG/DOPC-GUVs containing water-soluble fluorescent dye and loaded with smaller vesicles were investigated using confocal microscopy. The fluorescence intensity (FI) of the GUV membrane due to CF-R9 increased with time to a saturated value, then the FI of the membrane of the smaller vesicles inside the GUV increased without leakage of dye, indicating that the CF-R9 entered the GUV without pore formation. Rate of entry of CF-R9 was low. Based on these results we discuss the mechanism of the entry of Rn.

[1] Biochemistry,53,386,2014

2Pos151 低い pH が誘起する DOPS/MO 膜の液晶相からキュービック相への相転移に対する温度効果

The Effect of Temperature on the Low pH-Induced Lamellar to Bicontinuous Cubic Phase Transition in DOPS/MO

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Recently we revealed the kinetic pathway of the low pH-induced L to double-diamond cubic (Q_{II}^{D}) phase transition in dioleoylphosphatidylserine (DOPS)/monoolein (MO) [1]; at the initial step, the L_{α} phase was directly transformed into the hexagonal II (H_{II}) phase, and subsequently the H_{II} phase slowly converted into the Q_{II}^{D} phase. Here, we investigated the effect of temperature on this phase transition. The rate constant of the initial step increased with temperature, but that of the second step decreased or kept constant with temperature. Based on a theory, we analyzed these results and obtained information on the activation energies and the structure of activated state of each step of this phase transition.

[1] Langmuir, 30, 8131, 2014

2Pos152 Effects of Cholesterol on the Entry of Cell-Penetrating Peptide Transportan 10 (TP10) into a Single Vesicle of Lipid Membranes

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We demonstrated that a fluorescent probe-labeled TP10 (CF-TP10) entered single GUVs of DOPG/DOPC or DOPC membrane (1). Plasma membranes of eukaryotic cells contain high conc. of cholesterol, which affect physical properties of membrane. We investigated the effect of cholesterol on the entry of CF-TP10 into a single GUV of membranes containing high conc. of cholesterol. The result indicates that CF-TP10 entered single GUVs of these membranes before CF-TP10-induced pore formation, but higher conc. of CF-TP10 were required for the entry compared with the membrane without cholesterol. This result clearly shows the TP10 can translocate across membranes containing cholesterol. We discuss its mechanism based on others experimental results.

(1) Biochemistry 53, 386, 2014

2Pos153 アクチン骨格様の DNA ゲル薄膜で支持されたリポソームの構築

Liposomes with skeleton network of self-assembled DNA gel mimicking actin cortex

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Liposome is the most popular artificial cell. Although un-isotonic conditions cause collapse or shrink of liposomes, living cells are tolerant to such osmotic shocks. One of the reasons of the difference is the presence of cytoskeleton network of actin gel. To mimic such structure, we prepare liposomes with self-assembled gel of biopolymers. The gelation polymer we used is designed DNA molecule and forms network structure below a critical temperature. We encapsulate the DNA solution in microdroplets coated by a lipid layer, and spontaneously form a hollow capsule due to electrostatic interaction between the membrane and DNA gel. The gel networks of DNA raise stability of the liposomal membrane and prevent abrupt collapse under a hypertonic condition.

2Pos154 支持脂質二重膜へのプロテオリポソーム再構成過程の観察

Observation of reconstitution process of proteoliposome into supported lipid bilayer

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Supported lipid bilayer (SLB) is one of artificial cell membrane models, and reconstruction of membrane proteins such as ion channels is an important subject. We reconstituted proteoliposome (PL) obtained from Chinese hamster ovary cells expressing human ether-a-go-go-related gene channel into SLB, and observed the processes by using fluorescence microscope and atomic force microscope (AFM).

We prepared SLB consisting of phosphatidylcholine, phosphatidylethanolamine, cholesterol on a mica substrate by the vesicle fusion method. We added PL to SLB and incubated. AFM topographies showed that microdomains of ~0.5 nm in depth existed in the SLB. In-situ fluorescence observation revealed that fusion of PL proceeded at particular sites, which we attributed to the microdomains.

2Pos155 グラフェン酸化物が誘起する中性リン脂質膜からなる巨大単一膜ベシクルの形状変化

Graphene oxide induced structural transformation of single giant unilamellar vesicles of phosphatidylcholine membranes

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Graphene oxide (GO), which is single-atomic sheet of aromatic carbon modified with hydroxyl and carboxyl groups, have antibacterial activity. In this study, we investigated the interaction of GOs with PC-membranes using the single GUV method. During the addition of GO solution into the vicinity of a GUV, single GO flake was observed by phase contrast microscopy. We varied the area of the GO flakes in the range of approximately 10 to 10^2 μm^2 by sonication. In any case, GO flakes bound to PC-membrane of the GUV, causing the continuous decrease in the diameter of the GUV. On the basis of the detailed dependence of disruption process of GUVs on both the concentration and area of GO, we discuss the mechanism of the adhesion and interaction between GO and PC-membranes.

2Pos156 KcsA チャネルの細胞内ドメインによる不活性化への影響

The cytoplasmic domain regulates inactivation in the KcsA channel

Minako Hirano[1], Yukiko Onishi[2], Toru Ide[3] ([1]*GPI*, [2]*RIKEN, Qbic*, [3]*Okayama Univ.*)

Previously we revealed that the cytoplasmic domain (CPD) of the KcsA channel, a proton-activated K^+ channel, regulates channel gating by sensing pH. In the present study, we show that the electrostatic state of the CPD influences the rate of inactivation and conformation of the selectivity filter, which acts as the channel's inactivation gate. Current measurements revealed that protonated-mimicking mutations of the CPD not only removed inactivation, but also decreased K^+ selectivity. In addition, we demonstrate that large rearrangements of the CPD toward the membrane do not relate to inactivation. These results suggest that protonations of the CPD cause subtle rearrangements to induce inactivation.

2Pos157 微小な液滴接触膜の形成とイオンチャネル機能解析への応用
The formation of a contact bubble bilayer and its application to the functional analysis of ion channels

Masayuki Iwamoto, Shigetoshi Oiki (*Dept. Mol. Physiol. Biophys., Univ. Fukui Facult. Med. Sci.*)

In this study, we succeeded in the formation of a minuscule lipid bilayer (contact bubble bilayer, CBB) between water bubbles swelled out from the tip of glass pipettes in the lipid-containing oil. Because of smaller area of the CBB (ca. 80 μm^2) than the conventional planar lipid bilayers (>1000 μm^2), lower background noise on the electrical signal measurements was attained. The lipid composition of each leaflet of the CBB was controlled independently. A rapid perfusion system was also developed by introducing additional pressure-driven injection pipettes to the bubbles. We found that the solution inside the bubble (ca. 300 pL) was exchanged within 20 ms. Applications of the CBB for the characterization of ion channels were presented (Iwamoto & Oiki, *Sci. Rep.* 2015).

2Pos158 分子動力学で明らかにされる細菌機械受容チャネル MscL の N 末端領域の張力センサーとしての重要性
How Important is the N-Terminal Domain of Bacterial Mechanosensitive Channel MscL for Sensing Membrane Tension: Molecular Dynamics Study

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The bacterial mechanosensitive channel MscL is homopentamer of a subunit with two transmembrane inner and outer helices. The major issue of MscL is to understand the gating mechanism driven by tension in the membrane, however, it remains unclear which amino acids sense membrane tension and how the sensed force induces channel opening. Our MD study reveals that Phe78 showed exceptionally strong interaction with lipids among amino acids in outer helices facing the bilayer. Thus Phe78 was concluded to be the major tension sensor. In this study, we assessed how important of the N-terminal (S1) helices running parallel to the cytoplasmic membrane for sensing membrane tension. As a result, the some amino acids in S1 sense membrane tension as much as Phe78.

2Pos159 心筋 Na/Ca 交換の PMCA とカルシウムホールによる調節
Regulation of cardiac Na/Ca exchanger by PMCA via "calcium holes"

Takao Shioya (*Dept. Physiol. Fac. Med. Saga Univ.*)

Heart cells have plasma membrane Ca-ATPase (PMCA) and Na/Ca exchanger (NCX) on their sarcolemmal membrane. However, actual contribution of PMCA to the Ca extrusion has long been questioned. Here I examined the role of PMCA by recording whole-cell NCX current from heart cells. 1) Selective inhibition of PMCA enhanced the amplitude of the NCX current. 2) PMCA inhibition increased the local [Ca]i level around the NCX molecules, without changing the global [Ca]i. 3) Computer simulation predicted the development of a Ca-deficient intracellular nanodomain, or a “calcium hole”, around the operating PMCA molecule. Hence, I conclude that cardiac PMCA operates to regulate the nearby NCX, and potentially other sarcolemmal proteins, by creating a local "calcium hole" around it.

2Pos160 細胞-GUV 電気融合による μm スケールの人工物導入について
Electrofusion of Cell-GUV enables um-sized artificial objects transfer into live cells

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Introduction of artificial objects into live cells is an important topic in biotechnology. We have developed the novel method for introducing artificial objects ranging in size from 10nm to 1um into live cells, by using electrofusion with an artificial giant unilamellar vesicle (GUV, liposome). Dispersed cells and GUVs were placed into an electro chamber, and were exposed to an AC field to align cells-GUVs, and were also exposed DC pulse to induce transient electrofusion. Delivering magnetic beads, DNA origamis, plasmids were implemented. The transfer efficiency was evaluated by using fluorescence microscopy and flow cytometry. Based on these results, we believe that the method will be used for elucidation of cell mechanisms and even creation of artificial cells.

2Pos161 コレラ菌全走化性受容体ホモログの Che システム帰属
Che system assignment of all chemoreceptor homologs in *Vibrio cholerae*

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Vibrio cholerae has three chemotaxis-related signaling systems, among which only system II is essential for chemotaxis. The bacterium also has 45 chemoreceptor homologs, also known as to MCP-like proteins (MLPs), but which system they belong to has been largely unknown. Each Che system contains a homolog of CheR which catalyzes methylation of MLPs. For system assignment of MLPs, we co-expressed each MLP with each CheR homolog in *Escherichia coli*. About half of the MLPs were methylated by CheR2, suggesting the bacterium has ability to show taxis to various stimuli. CheR1 and CheR3 methylated about one quarter and a couple of MLPs, respectively. The rest was not methylated by any CheR homolog, and therefore was assigned to Che systems by informatic analyses.

2Pos162 新規光応答性 CaMKII の単一スパイン内光操作
Optogenetic manipulation of photo-activatable CaMKIIα in individual dendritic spines of neuron

Akihiro Shibata[1,2], Hideji Murakoshi[1] ([1]*National Institute for Physiological Science*, [2]*JSPS Research Fellow*)

CaMKII plays central roles in synaptic plasticity including long-term potentiation (LTP), which underlies some forms of learning and memory. Here we developed a photo-activatable CaMKIIα (paCaMKIIα) by using light oxygen voltage (LOV) domain. Blue light or 2-photon excitation can induce the conformational change of paCaMKIIα from the closed form (inactive) to the open form (active). The activation of paCaMKIIα in a single dendritic spine of neuron revealed that the CaMKII activation is sufficient to trigger the structural plasticity. Moreover, the activation of paCaMKIIα induced the recruitment of AMPA receptors into the plasma membrane of spines, most likely inducing long-term potentiation. Thus, paCaMKIIα is useful tool for the study of synaptic plasticity.

2Pos163 ナメクジ嗅覚中枢の培養ニューロンにおける神経振動ネットワークの再形成とアセチルコリン／ヒスタミンによる自発的神経活動の調節

Oscillatory network formation and cholinergic/histaminergic activity in the cultured olfactory neurons in the slug

Suguru Kobayashi, Asuka Kobayashi (*Kagawa Schl Pharmaceut Sci, Tokushima Bunri Univ*)

Synchronous oscillatory activity in a laminar structure is common in the olfactory system of both vertebrates and invertebrates. In the terrestrial slugs, periodic oscillation is recorded from the surface of the laminar structure of olfactory center (PC) and its frequency changes are suggested to encode the olfactory information and memory. We found the oscillatory network formation from dispersed cell culture of PC neuron. Calcium imaging for each PC neurons showed that AChE inhibitor or nicotine induced synchronous oscillatory activity and histamine increased spontaneous calcium transients without synchronous oscillation. It is suggested that acetylcholine and histamine can function as an excitatory transmitter in oscillatory network of cultured olfactory neuron.

2Pos164 海馬で合成される男性・女性ホルモンによる記憶シナプスの急性制御の解析

Rapid modulation of memory-related synapses by locally synthesized sex-hormones in the hippocampus

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Sex-steroids are synthesized locally in the hippocampus, center for learning and memory (Hojo et al., 2004, 2009). Analysis of confocal images by Spiso-3D software (Mukai et al., 2011) demonstrated that the application of testosterone (T), dihydrotestosterone (DHT) and estradiol (E2) rapidly increased the hippocampal spines (=postsynapses). Signaling pathways include synaptic AR and ER receptors > LIMK, MAPK, PKA, PKC > actin > spine increase. Selective kinase inhibitors suppressed the spine increase by T, DHT and E2. See (Hatanaka et al., 2015; Hasegawa et al., 2015; Murakami et al., 2015).

2Pos165 ミミズ非連合学習における 5-HT/NO/cGMP シグナルの役割

Role of 5-HT/NO/cGMP signaling cascade in non-associative learning of earthworm

Yoshiichiro Kitamura[1], Hitoshi Aonuma[2], Hiroto Ogawa[3], Kotaro Oka[4] ([1]*Dept Math Sci Phys, Kanto Gakuin Univ*, [2]*Res Inst Elect Sci, Hokkaido Univ*, [3]*Dept Biol Sci, Hokkaido Univ*, [4]*Dept Biosci Info, Keio Univ*)

Role of serotonin (5-HT) in non-associative learning in earthworm was investigated. Habituation by repeated tactile stimulus to the body wall in the earthworm is occurred assumedly due to via nitric oxide (NO)/cyclic GMP (cGMP) signaling, because both of NO and cGMP accelerated decrease of number of action potentials during repeated stimulus. Bath application of 5-HT also accelerated induction of habituation, and 5-HT antagonist conversely slowed induction of habituation. From our previous reports, 5-HT induced NO production in the ventral nervous system of the earthworm. Then, non-associative learning such as habituation in the earthworm by repeated tactile stimulus is supposedly induced by 5-HT/NO/cGMP signaling cascade.

2Pos166 柔らかい有機電極を用いる脳活動の同時多点測定

Multi-site recording of brain activity using flexible organic electrodes

Satoshi Watanabe, Hideyuki Takahashi, Keiichi Torimitsu (*Grad. Sch. Eng., Tohoku Univ.*)

Soft and biocompatible nature of organic material renders organic-based electrodes suitable for chronic multi-site recording of brain activity. We fabricated two types of electrode arrays using poly(3,4-ethylenedioxythiophene) p-toluenesulfonate (PEDOT-pTS) as conductive material. The first type had PEDOT-pTS-coated silk threads arranged on a patch of silk cloth. The second type had a thin layer of PEDOT-pTS/polyurethane formed on a silk film. Using these electrodes, we recorded spontaneous and stimulus-evoked activities in the embryonic chick brain with a high signal-to-noise ratio. These electrodes are expected to be suited for clinical applications such as diagnosis of neuropathological states.

2Pos167 2-AG 分解酵素モノアシルグリセロールリパーゼ欠損マウスにおける課題依存性学習障害

Task-specific impairment of hippocampus-dependent learning in mice deficient in monoacylglycerol lipase

Yasushi Kishimoto[1], Barbara Cagniard[2], Maya Yamazaki[3], Junko Nakayama[1], Kenji Sakimura[3], Yutaka Kirino[1], Masanobu Kano[2] ([1]*Department of Neurobiophysics, Kagawa School of Pharmaceutical Sciences, Tokushima Bunri University*, [2]*Department of Neurophysiology, Graduate School of Medicine, University of Tokyo*, [3]*Department of Cellular Neurobiology, Brain Research Institute, Niigata University*)

Growing evidence indicates that the endocannabinoid system is important for the learning and memory, however it is unclear which endocannabinoid plays a crucial role in it. Thus, we conducted a behavioral test battery in knockout (KO) mice deficient in monoacylglycerol lipase (MGL), the major hydrolyzing enzyme of 2-arachidonoylglycerol (2-AG). In the Morris water maze (MWM), MGL KO mice showed significantly faster memory extinction. In contrast, in the contextual fear conditioning, KO mice tended to show slower memory extinction. In the novel object recognition and water-finding tests, KO mice exhibited enhanced memory acquisition. These results indicate that 2-AG signaling is important for hippocampus-dependent learning, but its contribution is highly task-dependent.

2Pos168 X-ray Crystal Structure of TR: Implications for High Thermal Stability and High-Performance Optogenetic Availability

Takashi Tsukamoto[1], Kenji Mizutani[2], Megumi Takahashi[3], Taisuke Hasegawa[4], Naoki Hashimoto[2], Shigehiko Hayashi[4], Shin Takagi[3], Takeshi Murata[2], Yuki Sudo[1] ([1]*Okayama University*, [2]*Chiba University*, [3]*Nagoya University*, [4]*Kyoto University*)

We present the X-ray structure of Thermophilic rhodopsin (TR) at 2.8 Å resolution. TR is a light-driven proton pump discovered from Thermus thermophilus. Overall structure was similar to xanthorhodopsin, which is a proton pump having a carotenoid as a secondary chromophore. Comparing these proteins, we estimated possible mechanisms for the high thermal stability of TR together with the results of MD simulation. In addition, the high stability and proton pumping activity of TR allowed us to apply optogenetics. As a result, TR worked as a neural silencer and efficiently worked more than archaerhodopsin-3, which is the best rhodopsin-based neural silencer. On the basis of these results with other findings, we will discuss the potential of TR for future materials and tools.

2Pos169 Synechocystis sp. PCC 7509 由来の新規光駆動アニオンポンプの機能解析

Functional studies on a light-driven anion pump from Synechocystis sp. PCC 7509

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Recently, genomic analysis has revealed that retinal proteins exist in a variety of organisms. In this study, we performed functional study on a novel halide pumping retinal protein from Synechocystis sp. PCC 7509 living in freshwater. The protein was categorized into a new phylogenetic clade and showed maximal absorption at 520 nm. In addition, light-induced ion transport measurements revealed that it pumps halides such as Cl^- and Br^- as like as halorhodopsins. Notably, it pumps SO_4^{2-}ions, a large divalent anion. Now, we are investigating halide binding assay to determine the binding constant and spectroscopic analysis to estimate the photochemical properties. On the basis of these results, we will discuss the molecular mechanism of the halide transportation.

2Pos170 プロテオロドプシンのアルカリ性条件下における光誘起プロトン移動の pH 依存性

pH dependence of the photoinduced proton transfer in proteorhodopsin under alkaline conditions

Jun Tamogami[1], Keitaro Sato[2], Sukuna Kurokawa[2], Takumi Yamada[2], Toshifumi Nara[1], Makoto Demura[3], Takashi Kikukawa[3], Eiro Muneyuki[2], Naoki Kamo[3] ([1]*College Pharm. Sci., Matsuyama Univ.*, [2]*Grad. Sci and Eng., Chuo Univ.*, [3]*Fac. Adv. Life. Sci., Hokkaido Univ.*)

Proteorhodopsin (PR) is a light-driven proton pump from marine eubacteria. In this study, we investigated the pH dependence of the photoinduced proton transfer in phospholipid-reconstituted PR under alkaline conditions. The photoelectrochemical measurements with an indium-tin oxide transparent electrode and thin polymer film (Lumirror) revealed that the pH-dependent reverse of the sequence and direction of the photoinduced proton transfer occurs at alkaline pH. Moreover, this inversion of the proton movement occurred with the formation of a blue-shifted photoproduct, which was termed as the M-alkali intermediate (M_a). Based on these observations, an expected photocycle and proton translocation model in PR was proposed.

2Pos171 アフリカツメガエル由来(6-4)光回復酵素の変異解析

Mutational analyses of Xenopus laevis (6-4) photolyase

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(6-4) Photolyases ((6-4) PLs) are DNA repair enzymes that selectively repair the (6-4) photoproducts, utilizing the blue light. Recently, it was suggested that this repair reaction was successive two-photon process including the long-lived intermediate. Besides, some amino acid residues in (6-4) PL are highly conserved over species, and they would be the important factors in the repair reaction. In this study, mutational studies indicated that R410 was important in recognition of the DNA lesion, and that H354, H358 and Y412 residues were involved in the repair reaction. Interestingly, some of the mutants yielded the repaired products under the high intensity light irradiation. These results would help to understand the function of the residues.

2Pos172 量子化学計算を用いたプロテオロドプシン L105 変異体の波長シフトの解析

Theoretical analysis of the color-tuning mechanism of mutations at Leu105 in Green-Light Absorbing Proteorhodopsin

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Green-absorbing proteorhodopsin (GPR) is a light-driven proton pump found in proteobacteria. Mutations at L105 close to the retinal chromophore in GPR cause significant spectral shift. However, the color-tuning mechanism still remains unclear. In this study, we calculated the absorption maximum for the wild type GPR and its several L105 mutants using the INDO/S-CIS and QM/MM TD-DFT methods. The results indicated that the electrostatic effect between negative amino acids and protonated Schiff base cause a spectral blue-shift. Furthermore, the results suggest that the electron correlation effect is important to reproduce the experimental absorption maximum change for the mutations to aromatic amino acids.

2Pos173 Photoactive Yellow Protein におけるアルギニン 52 のプロトン化状態

Protonation State of Arginine 52 in Photoactive Yellow Protein

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We revealed R52 takes an electronic neutral form in the dark state (DS) of Photoactive Yellow Protein (PYP). In order to confirm the unusual protonation state of R52, we carried out IR experiments. Double difference IR spectrum of PYP_L minus DS between ^{15}N and ^{14N} -PYPs is composed of two negative and one positive major peaks. On the other hand, double difference spectrum in D_2O is composed of a pair of negative and positive peaks. DFT calculation was performed to interpret the IR bands. The number of the observed peaks in DS and PYP_L can be reproduced only by assuming that R52 takes an electronic neutral form at DS and a protonated form at PYP_L, respectively. These results reveal that the pK_a should be increased upon the PYP_L formation.

2Pos174 *Rhodobacter capsulatus* 由来 Photoactive Yellow Protein の相互作用における β-scaffold 部位の役割

Analysis of interaction sites on β-scaffold region of *Rhodobacter capsulatus* Photoactive Yellow Protein

Yoichi Yamazaki, Atsuhiro Kawamura, Mikio Kataoka, Hironari Kamikubo (*Materials Science NAIST*)

Photoactive Yellow Protein (PYP) is a blue light receptor. We identified the light dependent interaction protein of PYP named PBP from *Rhodobacter capsulatus* (Rc). Interaction mechanism of Rc-PYP and PBP has not been clarified yet. Crystal structure of Rc-PYP with a detergent molecule as a crystallization additive showed detergent molecule binding at the4-5 loop region of Rc-PYP. From this structure, β4-5 loop was thought as an interaction surface. To clarify this we verified interaction ability of β4-5 loop substituted Rc-PYP. Loop substitution affected Rc-PYP visible absorption spectrum but it kept interaction ability. However more extended substitution on b-scaffold region lost interaction. The β-scaffold region has key role for interaction of Rc-PYP.

2Pos175 ナトリウムポンプロドプシンの低温赤外分光研究
Low-temperature FTIR Study of Sodium Pumping Rhodopsin

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Krokinobacter eikastus rhodospin 2 (KR2) was discovered in 2013 as be the first light-driven sodium pumping rhodopsin. Until then, it was believed that cations cannot be transported by rhodopsin, because the protonated Schiff base is located within the ion-conducting pathway. Based on the structure and function study of KR2, sodium transport mechanism has been proposed.
In this study, we applied light-induced low temperature FTIR spectroscopy between dark state and each intermediate. From the FTIR spectra of unlabeled, ^{15}N Lys and Arg labeled proteins, we will discuss the detailed mechanism of sodium pump.

2Pos176 光駆動ナトリウムポンプ KR2 のポンプスイッチにおける Asn112 の役割
Role of Asn112 for transport activity by a light-driven sodium ion pump

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Nature created two kinds of light-driven ion pumps, outward proton and inward chloride pumps. In addition, we found a light-driven outward sodium pump, KR2, recently. KR2 has a conserved NDQ (N112, D116 and Q123) motif and pumps H^+ in KCl solution. The transport mechanism is intriguing, and in this study, we focused Asn112, which is involved in the hydrogen-bonding network in the recent structure of KR2.
We expressed mutants of N112 in *E. coli* and measured light induced pH change. N112D mutants still retained Na^+ transport activities, but N112A mutants lost Na^+ transport activities. We will discuss the role of N112 based on the present analysis including other N112 mutants.

2Pos177 部位特異的変異体を用いた bZIP モジュールである Photozipper の戻り反応の評価
Dark regeneration kinetics of site-directed mutants of bZIP module, Photozipper

Yuki Yabe, osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

Photozipper (PZ) has a basic leucine zipper domain and a light-oxygen-voltage-sensing (LOV) domain of aureochrome-1 and dimerizes upon illumination. The photoexcited PZ underwent the dark-regeneration with a half-life-time of 7.4±0.2 min at 25℃. The number of photoexcited PZ depends on the intensity of the incident light and the life-time of photoexcited state. In this study, we attempted to modulate the kinetics of dark-regeneration by replacing amino acid residues within the LOV core. Substitution of V300M showed 7-fold faster regeneration course, although V220I has 2.5-fold longer life-time than that of wild-type. The light-induced dimerization of these mutant PZs was investigated by dynamic light scattering and size exclusion chromatography measurements.

2Pos178 酸素発生複合体におけるＳ４状態での３重項酸素発生に関する理論的研究
Theoretical study on evolution of triplet oxygen molecule at the S4 state of oxygen evolution complex

Yasunori Yoshioka[1], Tomoya Ichino[2] ([1]*Mie University*, [2]*Hokkaido University*)

During the catalysis of water oxidation in nature, the oxygen evolving complex (OEC) passes through five oxidation states (S0 - S4). The mechanism of oxygen evolution at the S4 state of OEC is still unsolved. We have previously shown that Mn(III)-OOH with the oxidation state of [2Mn(III), 2Mn(IV)] is formed as a precursor of oxygen evolution at the initial stage of S4 state. In this work, we will show that Mn(III)-OO with the oxidation state of [2Mn(III), 2Mn(IV)] is formed from Mn(III)-OOH through the energy barrier less than 3.0 kcal/mol. The release of OO from Mn(III)-OO proceeds through the lower energy path than Mn(III)-OOH to give triplet oxygen molecule and [Mn(II), Mn(III), 2Mn(IV)].

2Pos179 好熱性紅色光合成細菌 Thermochromatium tepidum 由来 LH1-RC 複合体におけるキノン分子の検出
Detection of quinone molecules in the LH1-RC complex from the thermophilic purple photosynthetic bacterium Thermochromatium tepidum

Mari Matsuzaki[1], Yuki Yura[1], Takashi Ohno[1], Seiu Otomo[2], Yukihiro Kimura[1] ([1]*Grad. Sch. Agri. Sci., Kobe Univ.*, [2]*Fac. Sci., Ibaraki Univ.*)

In purple photosynthetic bacteria, a light-harvesting 1 (LH1) associates with a reaction center (RC) to form a LH1-RC complex. The LH1 complex is comprised of 14-16 αβ-subunits arranged surrounding the RC with or without a gap, which is thought to be involved in a quinone transport. The recent crystallographic structure of the LH1-RC complex from thermophilic Tch. tepidum revealed the oval LH1 shape without the gap, and the presence of the quinone gate in the transmembrane region of the αβ-subunit. In the present study, the behavior of quinone molecules upon a photosynthetic charge separation of the Tch. tepidum LH1-RC complex was monitored by light-induced FTIR difference spectroscopy and differences between the thermophile and other species were discussed.

2Pos180 NMR study of the interaction sites on the two Fd isoforms for photosynthetic protein complexes

Risa Mutoh[1], Yuko Misumi[1], Hisako Kubota-Kawai[2], Ryutaro Tokutsu[2], Takahisa Ikegami[3], Hippler Michael[4], Jun Minagawa[2], Genji Kurisu[1] ([1]*Inst. Prot. Res., Osaka Univ.*, [2]*National Inst. Basic Biol.*, [3]*Dep. Med. Life Sci., Yokohama City Univ.*, [4]*Inst. Plant Biol. and Biotech., Univ. of Munster*)

In chloroplast, Ferredoxin (Fd) is reduced by Photosystem I (PSI) and oxidized by Fd-$NADP^+$ reductase (FNR). Green alga possesses two Fd isoforms, Fd1 and Fd2, implying isoform specific involvement in the formation of NADPH or ATP, respectively. However, the structural basis for the isoform specific electron transfer is still elusive. In this study, we performed NMR analysis to determine the interaction sites on Fd1 with FNR and PSI for NADPH production, and are trying to detect the sites on Fd2 with cyclic electron flow (CEF) supercomplex for ATP synthesis, composed of PSI, Cyt b_6f, light-harvesting complexes, FNR and other membrane proteins. Now, we can show the interaction sites on Fd for single FNR and PSI are at lease partly distinct.

2Pos181 Fluorescence spectroscopy of single Photosystem I at liquid nitrogen temperatures

Ting Du (*Tohoku University*)

We conducted the single molecule fluorescence spectroscopy of photosystem I (PSI) trimers purified from cyanobacteria using a novel cryogenic microscope. We found temporal fluctuations of fluorescence intensity of single PSI at 83 K. This blinking has not been reported in preceding studies done at 1.4 K. The blinking of single PSI at 83 K may be due to the dynamic conformation switching. We assume that in the dark sate the fluorescence is quenched by P700+ accumulated under the present experimental condition. We found that the blinkings occur more frequently when the laser power is increased. This suggested that the conformation changes occur mainly in the excited state. We will also discuss observed correlation between the fluorescence intensity and peak wavelength.

2Pos182 励起子緩和過程の摂動論に対する新たな試み
New approach of perturbative study in exciton relaxation process

Akihiro Kimura (*Department of Physics, Graduate School of Science, Nagoya University*)

Recently, we constructed variational master equation for excitation energy transfer (EET) in the photosynthetic antenna systems. However, in the intermediate coupling case, there were quantitative disagreements in the stronger EET coupling case.
To understand the cause of disagreement, we try to construct general perturbative theory of exciton relaxation process.
In order to obtain more quantitatively correct result of the phenomenon, we apply renormalization method to the ordinary perturbation theory. At the poster presentation, we will show the analytical final form, compare our previous study, and discuss them.

2Pos183 ENDOR studies on biochemical modification on calcium site of the Mn cluster in photosystem II

Hiroki Nagashima[1], Yoshiki Nakajima[2], Jian-Ren Shen[2], Hiroyuki Mino[1] ([1]*Grad. Sch, Sci., Nagoya Univ.*, [2]*Grad. Sch. Nat. Sci. and Tech./Fac. Sci., Okayama Univ.,*)

Mn cluster in photosystem II is a catalyst of water splitting and oxygen evolution in the oxygenic photosynthesis. The Mn cluster consists of four Mn, five O and one Ca. Oxygen evolving activity is lost by the Ca-depletion and recovered by reconstitutions of Ca or Sr ions. The role of Ca ion is still unclear. ENDOR is a powerful method to detect protons near the Mn cluster. In this study, we applied ENDOR to the Ca-depleted and Sr-substituted Mn cluster. ENDOR spectrum of Ca-depletion demonstrated the cutting of important hydrogen-bond near the Ca site. ENDOR spectrum of Sr-constituted PS II is similar to untreated PS II. These ENDOR results indicated that the Ca depletion inhibited efficient electron transfer and proton release in the water oxidation steps.

2Pos184 プリオンペプチドと二価金属錯体結合によってひきおこされるレドックス不均衡
Redox imbalance induced by coordination of metals in prion peptide

Shinnosuke Kondo, Masahiro Yagi, Wakako Hiraoka (*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*)

Prion protein (PrP) has several Cu^{2+}-binding sites, and its misfolding leads to neurodegenerative diseases. Physiological function of PrP is still unknown, however its redox and metal-reservoir potentials are considered to be significant.
In this report, we investigated the reactivity of metal-binding PrP with reactive oxygen species. We used the octapeptide (PHGGGWGQ) as Cu^{2+}-binding site, which was the octarepeat region of human PrP. CD analysis showed that the binding process of octapeptide-Cu^{2+} was reversible and that other divalent ions (Ni^{2+}, Co^{2+}, Zn^{2+}, and Mn^{2+}) were partially substitute for Cu^{2+}. ESR-spin trapping revealed that the metal-binding octapeptide converted superoxide into hydroxyl radical.

2Pos185 超音波による CMNB ケージ基解離と脂肪酸分解の比較
Comparison between CMNB-caged moiety and fatty acid on molecular scission induced by ultrasound

Kengo Takei[1], Haruko Koura[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Master. & Human Env. Sci., Shonan Inst. of Tech.*)

Ultrasound-induced excitation and scission of molecules is the first process of sonodynamic therapy. We have been tring to utilize ultrasound in the activation of caged compounds for drug delivery system (DDS). To achieve the safe and effective treatment using ultrasound, it is necessary to estimate both the effect of DDS and the damage of cell components. We used fluorescein bis-(5-carboxymethoxy-2-nitrobenzyl) ether (CMNB-fluorescein) as a caging model, from which fluorescein can be released after activation. Fatty acid was employed as the damage model of cell components. We will report the scission efficiency of molecules when CMNB-fluorescein and fatty acids were irradiated with ultrasound from 28 kHz to 5 MHz.

2Pos186 Self-Emergent Cell-Sized Microsphere Entrapping DNA in a Crowding Binary Polymer Solution

Naoki Nakatani[1], Kanta Tsumoto[2], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life Medical Sci., Doshisha Univ.*, [2]*Grad. Sch. Engineering, Mie Univ.*)

Living cells on the earth maintain their lives by utilizing micro-compartment entrapping essential biomolecular devices including DNA. Currently, such micro-compartmentalization has been studied by focusing on the structure of phospholipid membrane. On the contrary, a few studies have argued the origin of compartmented living cells is attributable to the spontaneous micro-segregation in a crowding environment with macromolecules. In the present paper, we have examined the aqueous solution with crowding binary polymer solution as the possible origin on the micro compartmentalization of primitive life. We found DNA molecules are specifically entrapped within the inner portion of cell-sized spheres rich in bulky polymers after segregation of the binary polymer solution.

2Pos187 **Culture-independent method for identifying microbial enzyme-encoding genes based on activity-driven single cell genomics**

Kazuki Nakamura[1], Ryo Iizuka[1], Takao Yoshida[2], Yuji Hatada[2], Yoshihiro Takaki[2], Shinro Nishi[2], Ayaka Iguchi[3], Dong Hyun Yoon[3], Tetsushi Sekiguchi[3], Shuichi Shoji[3], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*JAMSTEC*, [3]*Major in Nanosci. and Nanoeng., Waseda Univ.*)

We present a simple method for identifying enzyme-encoding genes from environmental microbes in a culture-independent manner. The method is based on activity-driven single cell genomics, which is focused on microbial cells showing desired enzymatic activities. First, environmental microbes are encapsulated at a single-cell level in water-in-oil droplets with the fluorogenic substrate for the target enzyme to screen the droplets containing a fluorescent microbe, which exhibits the desired enzymatic activity. Second, microbial cells are subjected to whole genome amplification. Finally, the amplified genomes are sequenced to identify genes encoding the target enzymes. Using this method, we obtained novel β-glucosidase genes from uncultured bacteria in marine samples.

2Pos188 **ヒト間期核の全ゲノム動力学シミュレーション**
Genome-wide chromatin dynamics simulation of human interphase nucleus

Shin Fujishiro, Naoko Tokuda, Masaki Sasai (*Grad. Sch. Eng., Nagoya Univ.*)

Eukaryotic genome is packaged in cell nucleus as chromatins. Increasing evidence suggests that the three-dimensional (3D) organization and dynamics of chromatins are closely related to gene regulation in interphase nucleus. Although several simulation studies have been made to investigate the 3D structure of chromatin, current understanding is mostly limited to local and static features of genome. So to obtain a physical picture of large-scale chromatin dynamics and gene regulation, we investigate the genome-wide chromatin dynamics by developing a new simulation model. In this model chromosomes are probabilistically anchored to nuclear bodies and locally looped to reproduce interaction frequencies observed in a Hi-C experiment.

2Pos189 **リガンド結合による構造変化がもたらす STING シグナル伝達系への影響**
Ligand-induced conformational changes in STING are essential for its signal transduction

Yuko Tsuchiya[1], Kenji Mizuguchi[2] ([1]*Institute for Protein Research*, [2]*National institutes of Biomedical Innovation, Health and Nutrition*)

STING (stimulator of interferon gene) is an essential adaptor protein in innate immunity. It senses cytosolic DNA and induces type I interferon (IFN) production when a cell is infected. A better understanding of the downstream signaling mechanism, triggered by ligand binding, is essential for the development of anti-cancer drugs and vaccine adjuvants. In this study, we performed molecular dynamics simulations of human STING in ligand-bound and unbound forms. Different motions were observed between the ligand-bound and unbound forms, and also between the structures bound by the most potent natural ligand and by a less potent one. Based on these findings, we attempt to elucidate the signaling mechanism of STING-dependent IFN production.

2Pos190 **ベクトルマッチアルゴリズムによるタンパク質—タンパク質ドッキングポーズを評価するための統計アミノ酸ペアポテンシャルの開発**
A statistical amino acid pair potential to re-rank protein-protein docking poses predicted by a vector match algorithm

Atsushi Hijikata, Masafumi Shionyu, Tsuyoshi Shirai (*Facl. Biosci., Nagahama Inst. Bio-Sci. Tech.*)

We have been developing a new method to predict the three-dimension structure of protein complexes using a vector match algorithm. Our method can generate better docking poses than those of the state of the art docking methods especially in case of conformation change observed between the ‘bound’ and ‘unbound’ forms. However, the ranking of docking poses still have a room for improvement. We hence attempt to develop a simple statistical amino acid pair potential, which based on the non-redundant protein complex structure data from the Protein Data Bank, for re-ranking the predicted docking poses. We will discuss the performance of using the simple statistical potential combined with our docking algorithm compared with those of the other docking methods.

2Pos191 **埋もれた極性残基の進化的保存**
Evolutionary conservation of buried polar residues

Matsuyuki Shirota[1,2,3] ([1]*Grad Sch Med, Tohoku Univ*, [2]*ToMMo, Tohoku Univ*, [3]*GSIS, Tohoku Univ*)

Polar residues buried in protein internal provide critical interactions in protein structure, but their relevance still remains to be elucidated due to their minor anomalous properties. To address this point, I comprehensively searched 7682 non-redundant protein structures for buried polar residues and analyzed their conservation in multiple sequence alignment. The conservation profiles revealed that buried polar residues are more constrained than buried hydrophobic ones in that two residues of the same charge are distinguished, whereas aliphatic residues are changeable with each other. These results suggest that buried polar residues play critical roles in determining the specificity of protein structures by participating in polar interactions.

2Pos192 **高い配列相同性を持ちながら異なる立体構造を持つタンパク質のアミノ酸配列と立体構造に基づく解析**
The differences in 3D structures of proteins with high sequence identity. Analyses of amino acid sequences and 3D structures

Kohei Ohnishi, Masanari Matsuoka, Masatake Sugita, Takeshi Kikuchi (*Ritsumeikan Univ.*)

How a protein folds into its native structure is not completely understood. To address the problem, we must know the relationship between the amino acid sequence and the 3D structure of a protein. In the case that the sequence identity is more than about 30%, it is generally accepted the topologies of two proteins are similar. However, newly some proteins that do not follow to this empirical rule have been artificially designed. Dealing with artificial proteins, GA and GB, having very different 3D structure while high sequence identity in this study. Comparing between their 3D structures on sequences is difficult problem. Here, in addition to using bioinformatics techniques, an analysis based on inter-residue average distance statistics is used to address the problem.

2Pos193 機能未知スプライシングアイソフォームの機能予測
Function prediction of uncharacterized splicing isoforms

Masafumi Shionyu (*Fac. Bio-Sci., Nagahama Inst. Bio-Sci. Tech.*)

Although a large number of alternatively spliced isoforms (splicing isoforms) are known, isoform-specific functions are less investigated experimentally. Because sequence differences between splicing isoforms from the same gene tend to be small, many function prediction methods based on similarity to function-known proteins often fail to identify isoform-specific functions. Therefore, 3D structure-based method for predicting function of uncharacterized splicing isoforms must be valuable. I am developing function prediction pipeline for splicing isoforms based on a prediction method of small molecule-binding sites on protein structure using amino acid propensities of interfaces to small molecules. I will discuss the accuracy of prediction results from the pipeline.

2Pos194 リゾチームスーパーファミリータンパク質のフォールディングユニットの頑健性.タンパク質機能とフォールディング機構との関係
Robustness of folding units in lysozyme superfamily proteins during evolution, relationship between functions and folding mechanisms

Takuto Nakashima, Michirou Kabata (*Ritsumeikan Univ.*)

To elucidate evolutionary effect on the folding mechanisms of a protein is important. It is not clear what extent the sequence homology is required to guarantee the similarity of the folding units in homologues of a protein. This study tries to clarify the robustness of folding units and mechanisms of proteins in lysozyme family using sequence analysis with interresidue average distance statistics. As a result, it is suggested that the folding mechanisms are different among proteins with different functions even if these proteins belong same family. In addition, it is suggested that 3D structures of folding units are partially conserved in superfamily.

2Pos195 構造変化を伴う高分子の反応拡散系―構造と機能のクロストーク
Reaction-Diffusion Systems with Polymers Consisting of Mechanical Units: Crosstalk between the Structure and Function

Yuichi Togashi (*RcMcD, Hiroshima Univ.*)

There exist a variety of molecular machines working on DNA in the cell nucleus. The DNA (chromatin) structure may affect binding and function of such machines; in turn, the machine operation can alter the DNA structure. We previously constructed a simple reaction-diffusion model consisting of molecular machines; each machine was modeled as a particle with an internal state variable, on which the shape of the particle depends. We observed complex behavior such as segregation of active and inactive clusters. Using this framework, by connecting the particles by elastic strings, here we present a model to study the abovementioned crosstalk between the structure and function of active polymers. Relevance to actual chromatin structural dynamics will be also discussed.

2Pos196 ミドリゾウリムシの細胞内共生における共生藻の単純な維持機構
Maintenance of algal endosymbionts in *Paramecium bursaria*: a simple model based on population dynamics

Sosuke Iwai, Kenji Fujiwara, Takuro Tamura (*Faculty of Education, Hirosaki Univ.*)

Algal endosymbiosis, in which host organisms contain photosynthetic algae within their cells, is widely distributed in eukaryotes. To maintain a stable symbiotic relationship, number of endosymbionts in the host must be maintained at a constant level; however, the mechanisms for maintaining algal endosymbionts are still largely unknown. Here, we investigate the population dynamics of the unicellular protist *Paramecium bursaria* and its *Chlorella*-like algal endosymbiont, and propose that endosymbiont population size in *P. bursaria* is regulated not by direct mechanisms such as cell division coupling, but rather by an indirect mechanism based on their different growth properties. Our model may provide a basis for understanding the maintenance of algal endosymbionts.

2Pos197 遺伝子発現振動系のダイナミクス低減
Dynamical Reduction Approach reveals Oscillating Gene Expressions

Ikuhiro Yamaguchi[1], Yutaro Ogawa[2], Akihiko Akao[2], Yuki Shimono[2], Yasuhiko Jimbo[3], Kiyoshi Kotani[4] ([1]*Grad. Edu. Univ. Tokyo*, [2]*Front. Sci. Univ. Tokyo*, [3]*Eng. Univ. Tokyo*, [4]*RCAST Univ. Tokyo*)

Oscillating gene expressions caused by delayed negative feedback loops play important role in life on earth. The analysis of them, however, is intrinsically difficult because of their infinite dimensionality. Furthermore, interactions between many kinds of molecules including protein monomers, protein dimers, and mRNAs, make the phenomena more complex. In this study, we provide some analytical methods to reduce the degree of freedom and to reveal mechanism of oscillating gene expressions.

2Pos198 複雑反応ネットワークに埋め込まれた時間階層構造の解読
Deciphering timescale hierarchy encoded in complex reaction networks

Yutaka Nagahata[1], Satoshi Maeda[2], Hiroshi Teramoto[1,3], Chun-Biu Li[2,3], Takashi Horiyama[4], Tetsuya Taketsugu[2], Tamiki Komatsuzaki[1,3] ([1]*Graducate School of Life Science, Hokkaido Univ.*, [2]*Faculty of Science, Hokkaido Univ.*, [3]*Research Institute for Electronic Science, Hokkaido Univ.*, [4]*Graduate School of Science and Engineering, Saitama Univ.*)

Energy landscape is one of the versatile concepts for understanding hierarchical organization of microstates in simple molecules to clusters, biomolecules, and their aggregates. Tree representations such as disconnectivity graph have focused on capturing the organization of states that help us to understand, e.g., foldability of proteins. However, conformational changes in biomolecules occur over a wide range of timescales, whose variability is inherent to systems. In this work, by newly defining a series of transition states over a given reaction network, our tree can reveal the timescale hierarchical nature of reactions/structural transitions buried in the network, and in what timescale some basins merge into a superbasin.

2Pos199 Interdomain communication as the mechanism of correlation between circadian oscillation of KaiC phosphorylation and ATPase activity

Shota Hashimoto, Das Sumita, Masaki Sasai, Tomoki P. Terada (*Grad. Sch. Eng., Nagoya Univ.*)

By mixing three cyanobacterial proteins, KaiA, KaiB and KaiC with ATP, circadian rhythm of phosphorylation level of KaiC can be reconstituted in vitro. In addition, Terauchi and his colleagues have shown that the ATPase activity of KaiC in the absence of KaiA or KaiB correlates with the inverse of the period length of KaiC phosphorylation cycle in the presence of KaiA and KaiB. To elucidate the mechanism of this correlation, we constructed a structure-based model of KaiC single hexamer, assuming structural communication between the N-terminal CI domain and the C-terminal CII domain of KaiC. Stochastic simulation using this model has shown that the structural communication of KaiC monomer can give rise to the observed correlation.

2Pos200 人工 RNA 自己複製システムにおける宿主 RNA と寄生体 RNA の振動ダイナミクスと進化
Oscillating population dynamics and evolution of artificial Host-Parasite replication system in micro compartment

Norikazu Ichihashi[1], Yohsuke Bansho[2], Tetsuya Yomo[1,2] ([1]*Osaka Univ, IST*, [2]*Osaka Univ, FBS*)

In vitro reconstitution of living systems is a useful strategy to understand simple principles underlying complex behavior in nature. Here we performed a long replication of an in vitro system composed of two replicating RNA species, one of which (host RNA) replicate independently but the other (parasite RNA) replicates in a manner depending on the host RNA. Both RNAs continuously replicated with an oscillating population dynamics only when the system is compartmentalized, and the pattern of oscillation changes as a result of accumulation of mutations both in host and parasite. This result demonstrates that a complex ecological behavior, oscillation population dynamics and evolution, spontaneously appear with a simple set of self-replicating molecules and compartment.

2Pos201 微小液滴を用いた非線形化学反応間の相互作用の制御
Control of the interaction among nonlinear chemical reactions based on microdroplets

Tomoya Okuaki[1], Haruka Sugiura[1], Ryuji Kawano[2], Masahiro Takinoue[1] ([1]*Dept. Comput. Intell. Syst. Sci., Tokyo Tech*, [2]*Div. Biotech. & life Sci., TUAT*)

An interaction among cells plays an important role in collective behavior of cells. Especially, dynamic change of the interaction is important for information processing performed by living organisms. To date, the interaction has been studied using artificial cells as models of cells. Although interactions between artificial cells have been studied theoretically and experimentally, dynamic control of the interactions has not been realized yet. Here, we propose a droplet-microfluidic system that contains a nonlinear chemical reaction. The interaction between droplets is dynamically controlled by fusion and fission of droplets. We believe that our system will promote the understanding of the complex behavior exhibited by cells processing information.

2Pos202 細胞チップの電気生理学的特性評価のためのインピーダンス／細胞外電位計測システムの開発
Development of impedance measurement system for identification of cells

Kenji Matsuura[1], Fumimasa Nomura[2], Akihiro Hattori[1], Hiromi Kurotobi[2], Masao Odaka[1], Hyonchol Kim[1], Hideyuki Terazono[2], Kenji Yasuda[1,2] ([1]*Kanagawa Academy of Science and Technology*, [2]*Tokyo Medical and Dental Univ.*)

A single cell impedance measurement system for identification of cells has been developed.The impedance measurement showed (1) the increase of impedance of microelectrodes were correlated with its area decrease from 10-7 to 10-10 m2, (2) even the area of electrode decreased from 10-8 to 10-10 m2, the noise level of external field potential signals did not changed, and (3) the attachment of cells on microelectrode can be identified by significant increase of impedance. The results indicate the potential to evaluate the cell-to-electrode contact and degradation of electrodes, which might be used for the identification of different type of cells such as circulating tumor cells, which might occur epithelial-mesenchymal transition.

2Pos203 オンチップマルチイメージングセルシステムを用いた血液中の単一がん細胞を認識するためのイメージングバイオマーカー認識方法の評価
Evaluation of imaging biomarkers for identification of single cancer cells in blood by on-chip multi imaging cell system

Masao Odaka[1,2], Hyonchol Kim[1,2], Mathias Girault[1], Akihiro Hattori[1], Hideyuki Terazono[1,2], Kenji Matsuura[1], Fumimasa Nomura[2], Kenji Yasuda[1,2] ([1]*KAST*, [2]*IBB, TMDU*)

We have developed an on-chip multi imaging cell system to identify target cells based on their morphological characters, which we call “imaging biomarkers”. In this study, the bright-field (cell shape) and fluorescence (nucleus shape) images of (1) healthy rat blood, (2) cancer cell line, and (3) cancer cell-implanted rat blood were taken by the system, and imaging biomarkers were evaluated, respectively. In result, cancer cells were distinguished from healthy blood cells by using cellular and nucleus areas as markers. In addition, a port of cancer cells had awkward nucleus morphologies, which is also one useful marker for the identification. These results indicate that measurements of imaging biomarkers are practically applicable to identify cancer cells in blood.

2Pos204 FCS による HbA1c の計測
HbA1c measurement using FCS

Atsushi Matsuo, Yasutomo Nomura, Kyohei Maruyama, Kyohei Nakayama, Mayuka Chiba (*Maebashi Institute of Technology*)

FCS and FCCS were proposed as methods for diagnosing diabetes with HbA1c, because HPLC used previously has some problems such as high cost and time consuming. They are low running cost and not necessary cleaning. The first measured solution contained polyhapten of HbA1c model protein, diluted anti-HbA1c antibody (x10000) and fluorescence antibody of 1.4 × 10-8 M. FCS revealed the increase in diffusion time of fluorescent molecules, which suggested the immune complex formation of polyhapten and its fluorescent antibody within 1 hr. In the next step, we used HbA1c but not polyhapten. Now we plan to determine the optimal conditions of immunoreaction using FCCS. Although HPLC has these difficulties, it may be possible for FCS and FCCS to overcome them.

2Pos205 生細胞における 1 分子内在性 mRNA イメージングのためのアンチセンスプローブの設計と評価

Design and evaluation of potent antisense probes for imaging individual endogenous mRNA in live cells

Shunsuke Takeda[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharm. Sci., Univ. of Tokyo*, [2]*JST, PRESTO*)

Investigating gene-specific and dynamic behaviors of mRNA is important in understanding cell functions. To reveal those mRNA behaviors in living cells, we adopted fluorescence imaging of individual endogenous mRNA using antisense probes. For this purpose, it is necessary to establish a methodology for obtaining the probe with high binding rate. Here, we investigated the relationship between probe sequence and binding rate in living cells. As a result, we suggest a general strategy for designing potent antisense probes for target mRNA. By complying with the strategy, we were able to obtain high affinity probes effectively, which enabled to observe bright spots derived from individual endogenous mRNA. These results highlighted the advantage of our approach in mRNA study.

2Pos206 蛍光偏光相関分光法による蛍光タンパク質の回転拡散の研究

Study of rotational diffusion of fluorescent proteins using polarization dependent fluorescence correlation spectroscopy (pol-FCS)

Makoto Oura[1], Johtaro Yamamoto[2], Masataka Kinjo[2] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Advanced Life Sci., Hokkaido Univ.*)

Fluorescence correlation spectroscopy (FCS) is a powerful tool for analyzing diffusion of fluorescent molecule *in vitro* and *in vivo*. However, conventional FCS mainly detects fluorescent fluctuation signals originated from translational diffusion of the fluorescent molecule. On the other hand, fluctuation signals from rotational diffusion also could be detected using the fluorescence polarization changing at nano second time scale. This study reports, at the first time, the molecular size and viscosity dependency of rotational diffusion of EGFP oligomers using pol-FCS. Moreover, we also performed rotational diffusion measurement in gel that thought to form unique structure. The results indicate pol-FCS has high sensitivity to nano-environment of solutions and materials.

2Pos207 電子線の動電現象による単一接着性細胞への局所的な染色液導入の観察

Observation of Local Dye Inflow into Single Adherent Cells induced by Electrokinetic Phenomena of Electron Beam

Moto Yoshioka, Hiroki Miyazako, Akira Wagatsuma, Kunihiko Mabuchi, Takayuki Hoshino (*Grad. Sch. IST., Univ. Tokyo*)

Single-cell electroporation (SCEP) had been applied to various analyses of cell functions. However, local electroporation (EP) on cell membranes was difficult due to the low resolution. We proposed here a SCEP using a fine focused electron beam (EB). Since the EB could give electrokinetic phenomena focused in sub-micrometers, the fine EP could be performed on cell membranes. Using our method, we reported here the electrokinetically induced local EP for single adherent living cells. Observing transient response of Propidium Iodide (PI) dye flourescence, the EB-induced PI dye inflow into the cells and intracellular diffusion of PI dye were confirmed. Although the viability of the treated cells was still unclear, the local SCEP probability by our method was demonstrated.

2Pos208 結合速度と光感受性を改良した蛍光 ATP バイオセンサー

Improvement of binding speed and photostability of fluorescent ATP biosensor for extracellular ATP imaging

Hiromi Imamura, Sui Nishiyama, Ryuta Iwakiri, Akira Kakizuka (*Grad. Biostudies, Kyoto Univ.*)

Extracellular ATP signaling has been implicated in diverse biological processes. However, spatio-temporal behavior of extracellular ATP is not well understood because of the limited methods to monitor it. Since ATP released from cells will be rapidly cleared from the environment, released ATP should be detected with high temporal resolution. In this study, we engineered a genetically encoded fluorescent ATP biosensor, QUEEN, to improve binding rate by introducing a disulfide bond into the ATP binding domain. An apparent binding rate of the mutant QUEEN was approximately 100 ms. We also improved photostability of QUEEN by introducing a point mutation. This new ATP biosensor will allow us the high-speed imaging of extracellular ATP signaling.

2Pos209 高速 AFM による DNA ジャイレースのダイナミクスの直接観察

Direct observation of dynamic action in DNA gyrase by high-speed AFM

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DNA topoisomerases are molecular machines that solve the DNA's topological problems associated with DNA replication, transcription and recombination. The enzymes add or remove DNA supercoils by transiently breaking one DNA segment and transporting another segment through the break.

DNA gyrase, a heterotetramer composed of two A and two B subunits, is a unique topoisomerase that can introduce negative supercoils into DNA in an ATP-dependent manner. Although numerous structural and biochemical studies have been carried out, the detailed process remains elusive due to the lack of direct observation of the DNA-protein complex in dynamic action.

In this study, we used high-speed AFM to visualize directly the dynamic behavior of individual subunits within gyrase in solution.

2Pos210 電子顕微鏡画像処理及び画像解析のための Eos/PIONE の開発

Development of Eos and PIONE for Image Processing and Analysis of Electron Micrographs

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Three-dimensional electron microscopy is one of the most powerful techniques for elucidating structure of proteins and their complexes and/or cellular architecture. Image processing and analysis is essential for the technique. We have developed a system called Eos/PIONE. Eos is a platform for image processing analysis, and PIONE is a platform for process management to use heterogeneous computers paralelly and effectively. Eos has more than 400 tools, some of which were implemented with GPGPU. Furthermore automated GUI called zephyr, which is written in JavaScript and used under a browser, was also implemented. For PIONE, we have developed a front end for submitting jobs and data using 'Dropbox' or file uploading. We will here report their developmental progress.

2Pos211 ラスター画像相互相関分光法による生細胞内外来 DNA 分解活性の時空間的可視化

Raster image cross-correlation method for spatiotemporal visualization of intracellular degradation activities against exogenous DNAs

Akira Sasaki[1,2], Johtaro Yamamoto[3], Takashi Jin[2], Masataka Kinjo[3] ([1]*BMRI, AIST*, [2]*QBiC, Riken*, [3]*Faculty of Adv. Life Sci., Hokkaido Univ.*)

Reducing intracellular DNA degradation is critical to enhance the efficiency of gene therapy. Exogenous DNA incorporation into cells is strictly blocked by the intracellular nuclease activity. Raster image cross-correlation spectroscopy (ccRICS) are image-based correlation method. Here we performed spatiotemporal ccRICS analyses of fluorescent DNA distribution and directly monitored the process of exogenous DNA degradation in living cell. Such direct monitors of DNA degradation allow us to determine the fate of the exogenous DNA. On comparing the process, our study shows that cytoplasmic nuclease activity differs between cell lines. We propose that the difference of nuclease activity in cytoplasm dictates a different resistance to exogenous DNA incorporation.

2Pos212 Novel green fluorescent protein from Olindias formosa with exceptional pH stability

Hajime Shinoda[1], Yuanqing Ma[2], Tomoki Matsuda[1,3], Takeharu Nagai[1,3] ([1]*Grad. Sch. Eng., Univ. Osaka*, [2]*Univ. Western Sydney*, [3]*ISIR, Univ. Osaka*)

The world's most utilized green fluorescent protein, EGFP from a jelly fish, Aequorea victoria has enormously contributed as a fusion tag to visualize biological phenomena. However, loss of fluorescence in acidic environment due to the pH sensitivity (pKa = 6.0) has hampered the application of EGFP to imaging in acidic compartment such as lysosome and secretary granule (pH~5.5). Here, we report a world's first pH resistant GFP, mfGFP cloned from flower hat jelly fish, Olindias formosa. mfGFP is 2.4-fold brighter than EGFP and has excellent pH stability (pKa = 3.8). Furthermore, we developed an indicator for the protease that activates in acidic condition. We will demonstrate the application of mfGFP to bioimaging that has been impossible with conventional GFP.

2Pos213 赤外超解像顕微鏡法による毛髪 α-ケラチンの分子配向イメージング -振動モード毎の偏光依存性測定-

Orientation-sensitive IR super-resolution imaging of human hair α-keratins -Polarization dependency measurements-

Kohei Ushio, Yukihisa Watase, Masaaki Fujii, Makoto Sakai (*Tokyo Institute of Technology*)

Vibrational sum-frequency generation (VSFG) detected IR super-resolution microscope has a possibility to measure the orientation-sensitive IR image with sub-micrometer scale spatial resolution. In this study, we applied a VSFG-detected IR super-resolution microscope to the oblique sections (Cutting angle : α = 0-90 degree) of human hair which is formed by α-keratin fibers, and attempted IR imaging at the IR super-resolution in the 3-9 μm mid-IR region.

From the vibrational modes dependence (the amide I, III, CH str. and NH str.) of VSFG signals, it is concluded that α-keratins are well oriented along the longitudinal direction of the human hair. In the presentation, the results of the polarization dependence of VSFG imaging will be also reported in detail.

2Pos214 １分子イメージングによる転写伸長因子 NELF と DSIF のダイナミクス定量解析

Quantitative analysis of dynamics of negative elongation factor NELF and DSIF by single molecule imaging

Daichi Ikeda, Yuma Ito, Makio Tokunaga, Kumiko Sakata-Sogawa (*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*)

Negative elongation factors, NELF and DSIF regulate transcription by promoter-proximal pausing. Despite of the important role in transcription regulation, detailed dynamics of NELF and DSIF is still unclear. Aiming to elucidate the dynamic properties of these elongation factors in the living cells, we performed single molecule imaging and quantitative analysis. We constructed SNAP-tag fusion protein of NELF and DSIF and established cell-lines expressing EGFP-fusion protein of rpb1, the largest subunit of RNA polymerase II, together with SNAP-NELF or SNAP-DSIF. By controlling the concentration of fluorescent ligand to SNAP, we visualized the movements of single molecules of NELF and DSIF in the nucleus. We will discuss the results of quantitative analysis.

2Pos215 ストレス顆粒内一分子 mRNA の超解像イメージング

Super-resolution imaging of single mRNA in stress granules

Yuki Suzuki[1], Ko Sugawara[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. Pharma., Univ. Tokyo*, [2]*JST, PRESTO*)

During stress, cytoplasmic mRNAs aggregate and form stress granules (SGs), where they are remodeled for repression of translation. However, the mechanism of translational repression in SGs has been unknown because a detailed conformation, a distribution and dynamics of mRNAs in SGs remain unclear. We combined multicolor labeling of single mRNA and super-resolution microscopy to investigate the properties of single mRNA. We observed two spots in close proximity which were most likely two points on single mRNA in SGs in cells. We also investigated the detailed distribution and single-particle dynamics of mRNA in stressed cells to understand the mechanism of SG formation.

2Pos216 ディフュージョンマップ法を用いたタンパク質位相回復像の分類

Classification of phase-retrieved projection electron density maps of a protein using the diffusion-map method

Takashi Yoshidome[1], Oroguchi Tomotaka[2,3], Masayoshi Nakasako[2,3], Mitsunori Ikeguchi[4] ([1]*Department of Applied Physics, School of Engineering, Tohoku University, Japan*, [2]*Department of Physics, Faculty of Science and Technology, Keio University*, [3]*Research Infrastructure Group, Advanced Photon Technology Division, RIKEN Harima Institute, Japan*, [4]*Graduate School of Medical Life Science, Yokohama City University, Japan*)

Coherent X-ray diffraction imaging (CXDI) experiment enables us to visualize the structures of non-crystalline particles with micrometer to sub-micrometer dimensions. Using the X-ray free-electron laser (XFEL), two-dimensional diffraction patterns are collected in the diffraction-before-destruction scheme. In order for future applications of the experiment to proteins, we require an algorithm that can classify the data with respect to polymorphic structures of proteins arising from conformational dynamics. In the presentation, using the diffusion-map method, we classify projection electron density maps retrieved from the diffraction patterns. We will discuss how each image is classified and a possibility of the construction of three-dimensional structures of a protein.

2Pos217 ラスター画像相関分光法(RICS)による生細胞内の DNA 分解過程の時空間解析

Spatiotemporal analysis of exogenous DNA degradation in living cells by raster image correlation spectroscopy (RICS)

Takashi Horio[1], Johtaro Yamamoto[2], Akira Sasaki[3], Masataka Kinjo[2] ([1]*Lab. Mol. Cell Dynamics, Grad. Life Sci., Hokkaido Univ.*, [2]*Lab. Mol. Cell Dynamics, Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*Biomedical Research Inst., AIST.*)

Gene transfection technique is used in the wide research field such as biology, medicine and gene therapy. However, the exogenous DNA competes with degradation in cytoplasm during the transportation to nucleus and it makes decrease of gene transfer efficiency. The large part of this degradation mechanism have not been clarified yet. To reveal this mechanism, degradation process of the exogenous DNAs in living cells was monitored by diffusion map obtained by combination of RICS and the global fitting method. By using global fitting, the quantitative diffusion coefficient in RICS was obtained in wider range. The drastic increase of diffusion coefficient of DNAs in cytoplasm was observed but not in nucleus around ten minutes after DNAs were injected into living cells.

2Pos218 コヒーレント X 線回折イメージング法に向けたフーリエ変換ホログラフィー法による初期位相決定法の開発

Application of Fourier transform holography to initial phasing in coherent X-ray diffraction imaging

Yuki Takayama[1], Yayoi Inui[2], Yuki Sekiguchi[1,3], Sachihiro Matsunaga[2], Masayoshi Nakasako[1,3], Koji Yonekura[1] ([1]*RIKEN SPring-8 Center*, [2]*Sci. Tech., Tokyo Univ. Sci.*, [3]*Sci. Tech., Keio Univ.*)

Coherent X-ray diffraction imaging (CXDI) has revealed internal structures of whole biological cells/organelles at resolutions of several nm. In CXDI, oversampled diffraction patterns can be phased by combinational use of phase-retrieval and shape-estimation algorithms, and thereby yield projected electron density maps of samples. However, lack of low-resolution information due to the beamstop is likely to produce incorrect solutions. We have developed a new phasing method incorporating Fourier transform holography, which yields the unique sample shape. This method can dramatically improve convergence of the phase retrieval and we successfully reconstructed whole chloroplasts of red algae from diffraction pattern collected with an X-ray free electron laser.

2Pos219 血管内皮細胞の自発的管形成能力を利用した in vitro 組織培養システムの構築

In vitro perfused tissue culture system using spontaneous vascular formation of endothelial cell

Yuji Nashimoto[1,4], Akiko Nakamasu[2,4], Hisako Imamura[2,4], Hidetoshi Kotera[1], Koichi Nishiyama[3,4], Takashi Miura[2,4], Ryuji Yokokawa[1,4] ([1]*Kyoto University, Graduate School of Engineering*, [2]*Kyusyu university, Graduate School of Medical Sciences*, [3]*Kumamoto University, Graduate School of Medical Sciences*, [4]*JST, CREST*)

Due to lack of proper vascularization methods, the current tissue engineering has the problem in constructing complex 3D tissues. In this presentation, we proposed the in vitro perfused tissue culture system using spontaneous vascular formation of endothelial cell. The spheroid used as tissue model was cultured in the central channel of microfluidic device. The newly sprouting vessel was induced by natural angiogenesis manner from the two microchannel that adjoined the central channel.
As a results, the sprouting vessel was successfully migrated and contacted to the spheroid. Because this anastomosis methods was based on natural angiogenesis, this framework demonstrates potential for applicability to various tissue.

2Pos220 マイクロ波照射下での酵素反応の促進効果

Acceleration of Enzymatic Reaction under Microwave Irradiation

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We have studied the effect of microwave to protease reaction and rolling circle amplification (RCA). In this study, microwave cavity resonator was used. By irradiating variety output, we evaluated the relationship between the microwave power and the enzyme activity of the enzyme reactions. To control the temperature of reaction condition, the heat generation by microwave irradiated was cooled using a cooling system. Heat block was also used in the conventional method. As a result, the microwave irradiation, we revealed enzymatic reaction had been promoted. Also, the output of the microwave was different, the enzyme activity was changed. The effect of such microwave irradiation, was evaluated by examining the physical properties.

2Pos221 マイクロ凹形状構造によるサイズ選択的細胞回収への排除体積効果の寄与

Contribution of depletion effect to size-specific target cell purification using mirometer-sized concave structures

Hyonchol Kim[1,2], Hideyuki Terazono[2], Hiroyuki Takei[3], Akihiro Hattori[1], Kenji Matsuura[1], Fumimasa Nomura[2], Kenji Yasuda[2] ([1]*KAST*, [2]*Inst. Biomat. Bioeng., Tokyo Med. Dent. Univ.*, [3]*Facul. Life Sci., Toyo Univ.*)

A method for size-specific cell collection using micrometer-sized magnetic concave structures, "magcups", was developed. Magcups were fabricated by coating nickels on polystyrene microsphere templates and removing the templates. Cells were size-specifically captured to inner cavity of the magcups depending on the cavity sizes. To evaluate size-specific attraction mechanisms between targets and magcups, two different diameters of model target microbeads were mixed with magcups with and without nanoparticles (NPs). Beads having closer diameter of magcup cavity were selectively captured, and this tendency increased depending on the increase of NPs. These results suggest size-specific cell collection by micro-concave structure can be explained by typical depletion effect.

2Pos222 微生物（大腸菌）を使った化学物質センサーの開発

Development of chemical substance sensor by using micro organism (*E. coli*)

Hiroto Tanaka[1], Tadashi Matsukawa[1], Yasushi Naruse[2], Yukihiro Tominari[3], Masato Okada[4], Yoshiyuki Sowa[5], Ikuro Kawagishi[5], Hiroaki Kojima[1] ([1]*Bio ICT Lab, NICT*, [2]*CiNet, NICT*, [3]*Nano ICT, NICT*, [4]*Tokyo Univ.*, [5]*Hosei Univ.*)

Chemical substances (CS) affect behaviors of (micro) organisms. Then, the behaviors could be considered as encoded data of CS inputs (CSIs). Therefore, it is possible to speculate type of CSIs, if the behaviors can be decoded. However, most behaviors are stochastic and/or simplified, so that, it is difficult to decode the behaviors. Here, we report development of this decoding technique, allowing us to estimate CSIs. This technology paves the way to take advantage of the biological materials as CS sensors. We use *E. coli* as a CS detector, and statistically extract characteristic information of rotational behaviors, which is used as indicator of CSIs. Our decoding technique (CS sensing) based on bio-response and machinery learning could be applied to various fields.

2Pos223 リアルタイム局所化学刺激システムの開発と応用
Development and application of the real-time local chemical stimulation system

Masaru Kojima, Takahiro Motoyoshi, Mitsuhiro Horade, Kazuto Kamiyama, Yasushi Mae, Tatsuo Arai (*Grad. Sch. Eng. Sci., Osaka Univ.*)

For revealing detailed and localized biological information, a local environmental control technique is desired. For example, when analyzing the detailed and localized properties of single cells, this technique is important. In this study, we developed a local environmental chemical stimulation system. By using micro dual-pipettes, this system could control the local reagent concentration dynamically, freely and automatically. In this system, to reduce the diffusion of chemical solution, spout pipette and suction pipette were used. Furthermore, we apply this system to peel off certain area of confluent cells.

2Pos224 DNA origami を用いた直交性のある転写ナノデバイスの構築
Rational design of orthogonal gene transcription nano device on DNA origami

Takeya Masubuchi[1], Hisashi Tadakuma[2], Ryo Iizuka[3], Masayuki Endo[2], Takashi Funatsu[3], Hiroshi Sugiyama[2], Yoshie Harada[2], Takuya Ueda[1] ([1]*Grad. Sch. of Frontier Sci., The Univ. of Tokyo*, [2]*iCeMS, Univ. Kyoto*, [3]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

In the cell, gene expression is highly controlled. To create biologically inspired nanoscale device enabling the control of gene expression, we made hybrid nanomachine (T7-tile) using DNA origami tile as the skeletal structure and T7 RNA polymerase (T7-RNAP) as the functional module. T7-tile hybrid allowed us to evaluate the effects of intermolecular distance of enzyme (T7-RNAP) and substrate (target gene containing T7 promoter). We will show our recent achievements.

2Pos225 生体ナノポアと 3-way junction DNA を用いた 1 分子ロジックゲートの構築
Single molecule logic operations using 3-way junction DNA and biological nanopores

Masayuki Ohara, Ryuji Kawano (*Grad. Sch. Eng., Tokyo Univ. of Agr. and Tech.*)

Biological nanopores can detect single molecules electrically without the label and amplification of targets. In a previous study, we have demonstrated an electrical logic operation using DNA/RNA and nanopores in droplet system. However, this operation requires the stochastic analysis for obtaining the output. In this study, we tried to construct of the single molecule logic gate using 3-way junction DNA (3WJ) and an α-hemolysin nanopore. 3WJ cannot pass through the nanopore because it has a Y shaped stems consisted of two specific sites for enzyme reactions. In the system, when the two different enzymes cleave the 3WJ, the structure is changed and it can pass through the nanopore as an output. This system is promising the development of high-speed molecular computing.

2Pos226 近赤外レーザーによる安定な細胞集合体の構築
Construction of Stable Cellular Assembly with Optical Manipulation

Aoi Yoshida, Shu Hashimoto, Taeko Ohta, Kenichi Yoshikawa (*Doshisha University*)

We will report the successful construction of stable cellular assembly by using laser tweezers. Laser tweezers enable us to transfer a certain cell onto another cell under remote control without any mechanical devices directly interfering the cells. We adapt a crowding medium with hydrophilic polymer to assemble cells floating in aqueous medium. Interestingly, after the contact for a few minutes under laser twizzling in an aqueous medium containing suitable amount of hydrophilic polymer, the cells keep stable contact even in the absence of crowding polymer. Stable 3-D cellular assemblies may be constructed with such experimental procedure. Lastly, we will argue the future applicability of this methodology toward the development in tissue-engineering and biotechnology.

2Pos227 マイクロピペットによるリポソーム内への物質移入法の開発
Development of novel methods for introducing materials into liposomes with micropipettes

Shin Yoshida[1], Fumika Asari[2], Tomoyuki Kaneko[1,2] ([1]*LaRC, Grad. Sci. Eng., Hosei Univ.*, [2]*LaRC, Dept. Frontier Biosci., Hosei Univ.*)

Lipids are known to form liposomes by self-assemble in water solutions. Liposomes can encapsulate materials in their inside and have been used as a cell membrane model and a carrier of drugs. Although, materials could be encapsulated at the time of making liposomes, it was difficult to put materials into already making liposomes. To develop the novel methods for introducing materials into liposomes with micropipettes, we explored the fixation methods of liposomes to the bottom of chamber and the modified condition of micropipettes. To assess the utility of this method, we put fluorescent substance of the membrane non-permeable into liposomes. This novel method would be contributed to make the artificial cell and to evolve the drug delivery system.

2Pos228 生化学分析のための電子線による有機カチオン輸送制御
Transportation Control of Organic Cations Using an Electron-beam for Biochemical Analysis

Hiroki Miyazako[1,2], Kyoko Fujita[3,4], Nobuhumi Nakamura[3,4], Hiroyuki Ohno[3,4], Kunihiko Mabuchi[1], Takayuki Hoshino[1] ([1]*IPC, UTokyo*, [2]*JSPS Research Fellow*, [3]*Dept. Biotech., TUAT*, [4]*Grad. Sch. Eng., TUAT*)

Room-temperature ionic liquids (RTILs) have been applied to solvents for biochemical analysis in various ways, such as control of biochemical catalytic reactions or extraction of proteins. This study proposes a new control method for transportation of organic cations in RTILs using a focused electric field of an electron beam (EB). We demonstrated the concentration of quaternary ammonium cations by scanning EBs, and aggregation of gold nanoparticles by concentrating tetramethylammonium ions. These results indicate the possibility that the proposed method will achieve generation of gold nanoparticles or extraction of proteins at a nanometer scale, which will be applied for single-cell analysis and control.

2Pos229 ビリルビンを発色団とする蛍光タンパク質 UnaG のキロオプティカル特性

Chiroptical Properties of Bilirubin-based Fluorescent Protein UnaG

Togo Shimozawa, Yoh Shitashima, Miyabi Ishida, Toru Asahi (*Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*)

UnaG, a protein discovered in eel muscle, is a new class of fluorescent protein which utilizes its ligand of Bilirubin(BR), a heme metabolite, as its chromophore. BR is composed of two dipyrrinone moieties(DM) having each electric transition moment, and the conformation of BR cause chiroptical properties, such as Circular Dichroism(CD). Chiroptical properties provide information about electronic structure of chromphore, which is useful for understanding fluorescence mechanism. From the CD spectrum, we observed three peaks at 350, 430, and 500 nm as negative, positive, and negative peaks respectively, and assigned two peaks at 430 and 500 nm to excitonic interaction of two DMs in BR molecule, however, the peak at 350 nm suggested another excitonic interaction in UnaG-BR.

2Pos230 Measuring *Escherichia coli* flagellar filaments growth in real time using fluorescent microscopy

Ziyi Zhao[1], Chien-Jung Lo[2], Fan Bai[1] ([1]*Biodynamic Optical Imaging Center, Peking University, Beijing, China*, [2]*Department of Physics and Graduate Institute of Biophysics, National Central University, Jhongli, Taiwan*)

The bacterial flagellum protrudes from the cell body as a long, helical filament. These filaments grow at their distal ends with thousands of flagellins (FliC) , which are pumped out by the Type III flagellar export apparatus and travel through the central channel of the flagellum for assembly. In order to study this self-assembling process, we use the biarsenical dye FlAsH/tetracysteine labeling system to fluorescently tag FliC in live *Escherichia coli* cells and observe growth of flagellar filaments in real time. The tetracysteine tag is genetically incorporated, thus preserving endogenous control of FliC expression. This approach enables us to measure the flagellar growth rate and observe the dynamics of flagellin secretion.

3Pos001 Rap1B の構造とコンフォメーションスイッチ機構
The structure and conformational switching of Rap1B

Hiroki Noguchi, Sam-Yong Park, Jeremy Tame, **Satoru Unzai** (*Yokohama City University, Graduate School of Medical Life Science*)

Rap1B is a small GTPase involved in the regulation of numerous cellular processes. Like other members of the Ras family, the active GTP-bound form of Rap1B can bind to a large number of effector proteins and so transmit signals to downstream components of the signaling pathways. Unlike other Ras family proteins such as H-Ras and Rap2A, Rap1B crystallizes in an intermediate state when bound to a non-hydrolyzable GTP analog. Comparison with H-Ras and Rap2A reveals conservative mutations relative to Rap1B, distant from the bound nucleotide, which control how readily the protein may adopt the fully activated form. Crystallographic structures of mutant proteins show how these changes may influence the hydrogen bonding patterns of the key switch residues.

3Pos002 完全重水素化 HiPIP の構造と性質
Structure and characterization of perdeuterated HiPIP

Yuya Hanazono, Kazuki Takeda, Kunio Miki (*Grad. Sch. Sci., Kyoto Univ.*)

Hydrogen atoms, which account for about half number of the atoms in proteins, are important for understanding the function of proteins. Neutron crystallography is a useful approach to determine hydrogen positions directly. The use of perdeuterated crystals can enhance the signal to noise ratio because of avoiding the incoherent scattering of hydrogen atoms.
We previously reported the high-resolution (~0.7 Å) structures of high potential iron-sulfur protein (HiPIP) purified from *T. tepidum*. In this study, we constructed the expression system in *E. coli* of unlabeled and perdeuterated HiPIP. Moreover, we investigated the recombinant HiPIP from crystallographic and spectroscopic studies. The differences between the unlabeled and perdeuterated HiPIP will be discussed.

3Pos003 酸化ヌクレオチド分解酵素の高分解能 X 線回折
High resolution X-ray diffraction study of an enzyme for oxidative nucleotide processing

Keisuke Hirata, Teruya Nakamura, Mami Chirifu, Shinji Ikemizu, **Yuriko Yamagata** (*Grad. Sch. Pharm., Kumamoto Univ*)

Oxidized deoxynucleotides cause replicational errors because of their misincorporations into DNA. The MutT and related proteins prevent transversion mutations by hydrolyzing mutagenic oxidized nucleotides such as 8-oxo-dGTP and 2-oxo-dATP, and there is a difference in substrate specificities between them. E. coli MutT hydrolyzes 8-oxo-dGTP to 8-oxo-dGMP with extremely high substrate specificity. On the other hand, its human homolog has broad substrate specificity. In order to understand mechanisms of their substrate specificities, we solved the crystal structures of MutT and its homolog complexed with their substrates. For understanding of broad substrate specificity, the identification of hydrogen positions in the active site was tried.

3Pos004 Structural and functional studies on two kinds of perireceptor proteins (PRPs) working in chemoreception

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Two types of soluble proteins, odorant-binding protein (OBPs) and chemosensory protein (CSPs) were found in the chemosensory organ of animal and insects. They are supposed to transport odorants or pheromons to its receptor and/or modify chemoreception process. Thus, we named them perireceptor protein (PRP). The former are mainly composed of beta-strands and the latter, alpha-helixes. In order to elucidate the structural-functional relationships, we have expressed and purified functional forms of them. We analyzed the structural stability and functional changes of PRPs upon ligand binding using fluorescence binding assay and CD (circular dichroism) measurements.

3Pos005 ストマチンパートナータンパク質の結晶構造と多量体形成
Crystal structure of stomatin operon partner protein and formation of multimeric assembly

Hideshi Yokoyama[1], Ikuo Matsui[2] ([1]*Sch. of Pharm. Sci., Univ. of Shizuoka*, [2]*Biomedical Res. Inst., AIST*)

Stomatin / STOPP (stomatin operon partner protein) gene pairs are present in both archaeal and bacterial species, and their protein products may be involved in the quality control of membrane proteins. The crystal structure of the C-terminal soluble domain of STOPP PH1510 (1510-C) from the hyperthermophilic archaeon *Pyrococcus horikoshii* was determined at 2.4 Å resolution. The structure of 1510-C had a compact five-stranded beta-barrel fold known as an oligosaccharide/oligonucleotide-binding fold (OB-fold). According to crystal packing, 1510-C could assemble into multimers based on a dimer as a basic unit. This structure indicates that 1510-C functions as a scaffold protein to form the multimeric assembly of STOPP and stomatin.

3Pos006 分子動力学シミュレーションを用いたリガンド-レセプター間の最適解離経路を決定するための新規方法
A novel method to determine the optimal unbinding path between receptor and ligand using advanced molecular dynamics simulations

Gert-Jan Bekker[1,2], Narutoshi Kamiya[1], Haruki Nakamura[1] ([1]*IPR, Osaka Univ*, [2]*FBS, Osaka Univ*)

In order to accurately discriminate between valid and decoy docking poses between two proteins generated by rigid body docking, we are working on calculating the binding free energy by using MD simulations. For the first stage, we need to produce an unbinding path. Two popular methods are SMD and RAMD, however both have significant drawbacks in computational time and accuracy. We have developed a novel and efficient method, Iterative-Self-Optimizing Guided MD (isoGMD). First, the method roughly searches for disassociation paths in uniform directions by applying adaptive forces and velocities using GMD. Then isoGMD iteratively discards unlikely paths and optimizes the remaining paths using GMD. We have applied our method to various systems.

3Pos007 赤痢菌ニードル複合体の極低温電子顕微鏡による構造解析
CryoEM structural analysis of the needle complex from *Shigella flexneri*

Naoko Kajimiura[1], Fumiaki Makino[1], Takayuki Kato[1], Ariel Blocker[2], Keiichi Namba[1,3] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*Sch. of Cell. & Mol. Med., Univ. of Bristol*, [3]*RIKEN, QBiC*)

The needle complex (NC) is a large, membrane-spanning, core protein complex of the type III secretion system of pathogenic bacteria. CryoEM single particle image analysis has become a powerful tool, revealing the structure of the NC basal body, but high-resolution structural analysis of intact NC is difficult because stoichiometry of the membrane protein components of the export apparatus is not well defined and also due to structural heterogeneity in purified NC particles. We report refinement of the NC purification method from *Shigella* by using different detergents in different concentrations to improve intactness of the export apparatus. We will present cryoEM structure of intact NC and identification of some protein components in the structure.

3Pos008 低解像度密度マップへの複数のサブユニットのあてはめ計算 - 実験情報による拘束の利用 -
Multiple subunit fitting into a low resolution density map using experimental additional restraints

Takeshi Kawabata, Hirofumi Suzuki, Haruki Nakamura (*Inst. Prot.Res., Osaka Univ.*)

Single particle CryoEM typically produces 3D density maps of the macromolecular complex with low resolution. For understanding these maps, it is essential to fit atomic structure of individual subunits into the maps of the complex. We developed a program gmfit using Gaussian mixture model (GMM) to rapidly search many configurations. In order to enhance the performance, we try to include several additional experimental restraints, such as proximities between subunits. Restraints to be satisfied as follows: 1) overlaps with density map, 2) repulsions, 3) symmetries and 4) proximities between subunits. New energy functions of the additional restraints for GMM are designed. Two searching algorithms are developed and tested: segmentation & fitting and build-up method.

3Pos009 SAAP 力場を用いたシニョリンのレプリカ交換モンテカルロシミュレーション
Replica-exchange Monte Carlo simulation of chignolin using SAAP force field

Toshiki Suzuki, Taku Shimosato, Natsuki Babe, Toshiya Minezaki, Michio Iwaoka (*School Sci., Tokai Univ.*)

In our laboratory, we are developing a new force field, called the single amino acid potential (SAAP) force field, for molecular simulation of polypeptides. In the SAAP force field, the total potential energy is divided into the single amino acid potentials and the inter-amino acid interactions. In this study, in order to improve the accuracy and efficiency of structure sampling, the replica-exchange method was introduced to the SAAP simulation program. When the modified simulation program was applied to the molecular simulation of chignolin, the native β-turn structure was reasonably generated. The result suggested that the new SAAP simulation program is usable for molecular simulation of short polypeptides.

3Pos010 全原子 MD シミュレーションによるアラニンスキャニング変異を導入したアミロイド形成ペプチドの凝集解析
All atom molecular dynamics alanine scanning of amyloid-forming peptides in explicit solvent system

Satoshi Kosuda[1], Atsushi Suenaga[2], Gentaro Morimoto[3], Makoto Taiji[3], Yutaka Kuroda[1] ([1]*Dep. Biotech. Life Sci., Grad. Sch. Eng., TUAT.*, [2]*Molprof, AIST*, [3]*QBiC, RIKEN*)

We report an all-atom molecular dynamics (MD) investigation of amyloid forming peptides, NFGAILSS and its alanine-scanned mutants. We carried out 50 ns MD simulations of systems containing 27 peptides and approximately 30000 water molecules using MD-GRAPE3. All mutants formed cluster, but the wild-type sequence formed more beta structures than the Ala-scanned sequences. Especially the F2A mutation fully hindered the formation of beta structures as analyzed using DSSP, which was consistent with experimental results (Gazit et al., J. Bio. Chem., 2001). A detailed analysis of the beta structure formation suggested that interaction between 2Fs was important at the initial stage of beta bridge formation, but the bridges were later stabilized by hydrophobic residues.

3Pos011 エステル基を有する新規白金系複核制がん剤による DNA の高次構造と遺伝子活性への影響
Action of novel anticancer-active, dinuclear platinum complexes with ester group on the higher order structure and genomic activity of DNA

Yuta Shimizu[1], Yuko Yoshikawa[2], Takahiro Tsuchiya[3], Hiroki Yoneyama[4], Shinya Harusawa[4], Seiji Komeda[3], Tadayuki Imanaka[2], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Univ. Doshisha*, [2]*Univ. Ritsumeikan*, [3]*Univ. Suzuka Med. Sci.*, [4]*Univ. Osaka Pharm. Sci.*)

Cisplatin is well known as a potent anticancer drug. However, applicability of cisplatin has been limited due to severe side effects. As a new candidate toward useful anticancer therapy, a novel type of platinum compound, tetrazolato-bridged dinuclear platinum (II) complexes with ester group, have been developed recently. Here, we studied the effect of such novel drug on the higher order structure of DNA. We will show that these dinuclear Pt chemicals cause bridges between DNA segments, being much different from cisplatin. We have also examined the effect of these dinuclear complexes on the gene-expression. We will stress that studies on the higher-order structure of genome sized DNA provides useful insight toward the development of new potent cancer drugs.

3Pos012 Attempts at CA-type formal analysis of fibrous assembly of particles

Takashi Konno (*Univ. Fukui. Med. Mol. Physiol.*)

In the framework of 2D and 3D cellular automata (CA), transition rules leading to fibrously assembled "structures" were constructed and analyzed. The elements could represent proteins in an abstractive form. The analysis in high dimensional CA systems could naturally be unexhaustive, but careful choice of the CA transition rules gave valuable insights into the physical reality. The rules could also be translated into the "energy" term. "Fibrous" pattern of a state in the CA lattice could directly be regarded as "fibers", but more abstractive definitions of "structure" were also challenged. This study is an initial step towards elucidating hidden logics unconsiously employed for recognizing "structures" in daily and/or scientific life.

3Pos013 The first crystal structure of intact 3.8 MDa molluscan hemocyanin

Asuka Matsuno[1], Zuoqi Gai[2], Koji Kato[1,2], Sanae Kato[3], Takeshi Shimizu[4], Takeya Yoshioka[4], Hideki Kishimura[5], Tohru Terada[6], Yoshikazu Tanaka[1,2], Min Yao[1,2] ([1]*Grad. Schl. of Life Sci., Hokkaido Univ*, [2]*Facl. of Adv. Life Sci., Hokkaido Univ*, [3]*Asahikawa Med. Univ*, [4]*Hokkaido Ind. Tech. Cent*, [5]*Grad. Schl. of Fish. Sci*, [6]*Grad. Schl. of Agr. and Life Sci., The Univ. of Tokyo*)

Oxygen transportation is one of the most important process for living organisms. Molluscs and arthropods use hemocyanin, a copper containing protein, for this purpose. Mollscan hemocyanin is decamer or multi-decamer of approximately 400 kDa, which is the largest known protein. Because of the propensity of dissociation and extraordinary size, structural studies have relied mainly on electron microscopy. In this study, we determined the first crystal structure of intact hemocyanin, at 3.0Å resolution, composed of 31,679 amino acid residues, 80 Cu2O2 clusters, and 50 carbohydrates. Based on this atomic structure, we discuss assembly manner, functional relevance of carbohydrates, coordianation geometry around the oxygen binding sites, and evolutional implications.

3Pos014 二面角系基準振動解析プログラムの並列化と巨大分子への適用

Parallelization of the program for normal mode analysis in torsional angle space and application to supramolecules

Shigeru Endo[1], Hiroshi Wako[2] ([1]*Dept. Phys., Sch. Science, Kitasato Univ.*, [2]*Sch. Social Sciences, Waseda Univ.*)

We have developed a computer program that performs normal mode analysis (NMA) based on an elastic network model. Taking advantage of the relatively small number of degrees of freedom required to describe a molecular structure in dihedral angle space, we aimed to develop the program applicable to a full-atom system of any molecule less than 10^5 atoms stored within a single file in the Protein Data Bank (PDB). A generalized eigenvalue problem and the calculation of atomic fluctuations of every atom, which were time-consuming processes in the program, were parallelized with OpenMP. The results of NMA of supramolecules will be reported in the annual meeting.

3Pos015 Template based protein modeling using a target dependent template library

Kodai Takagi, George Chikenji, Yota Masuyama (*Grad. Sch. Eng., Nagoya Univ.*)

Template Based Modeling (TBM) is currently the most powerful protein structure prediction method. However, when only analogous proteins are available as templates, TBM may not produce a significant
prediction. To improve prediction accuracy for these targets, it is critically important to develop a method that correctly assesses the sequence-structure compatibility. For this purpose, we developed a new template based modeling method in which template structures are customized depending on the predicted secondary structure information of the target protein. This customization helps us appropriately assess the sequence-structure compatibility. In the presentation, the detailed description of the method and the results of benchmark tests will be presented.

3Pos016 レプリカ交換アンブレラサンプリング MD シミュレーションを用いたヒストン脱アセチル化酵素阻害剤の選択性の研究

Study for the selectivity of a histone deacetylase inhibitor using replica-exchange umbrella sampling MD simulation

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Histone deacetylases (HDACs) have a key role in an epigenetic regulation of gene expression by removing acetyl groups from lysine residues in histone tails, resulting in chromatin condensation. HDACs are thus targets of drugs for cancer, cardiovascular diseases or neurological disease. Recently, T247 was proposed as a selective inhibitor. Selectivity is important to reduce side effects.
We analyzed the inhibition mechanism of T247 for HDACs by using replica-exchange umbrella sampling (REUS) molecular dynamics (MD) simulation. This is powerfull method to obtain various conformation of system across energy barriers along a reaction coordinate. As a result of this simulation, we could reconstruct selectivity of T247.

3Pos017 Interaction of Two Subunits of D/L Hetero Dimer with Catalytic Subunits in Archaeal RNA polymerase: Insights from MD Simulations

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Using MD simulations and multimodal dynamic cross correlation (mDCC) analysis, we dissect the structural and functional roles of the domains 2 and 3 of D/L hetero dimer in the assembly and catalytic activity of euryarchaeal RNAP from *Thermococcus Kodakarensis*. The domain organization of DL dimer is almost conserved in archaeal-eukaryotic RNAP family except for substantial differences in domains 2 and 3, and its formation initiates RNAP assembly. MD simulation of DL hetero dimer and whole length RNAP indicates that several flexible loop regions in L subunit as well in domains 2 and 3 of D subunit is characteristically stabilized by interactions with the catalytic subunits. Several conserved residues that may play a key role in these interactions are also identified.

3Pos018 光活性化アデニル酸シクラーゼ合成酵素 PAC の活性化機構解明

Structural and functional insights into a photoactivated adenylyl cyclase

Mio Ohki[1], Kanako Sugiyama[1], Fumihiro Kawai[1], Shigeru Matsunaga[2], Naoya Shibayama[3], Mineo Iseki[4], Sam-Yong Park[1] ([1]*Grad. Sch. of Medi. Life Sci, Yokohama City Univ.*, [2]*Cent. Research Lab. Hamamatsu Photonics K.K.*, [3]*Dept. of Physiology Jichi Medi. Univ.*, [4]*Faculty of Pharma. Sci. Toho Univ.*)

Naturally occurring light sensor domains are able to control biological processes such as plant development and the behaviour of microbes by utilising the photochemical response of prosthetic flavins, and in recent years there has been growing interest in understanding and exploiting these proteins for synthetic biology. Here we describe crystal structures and the light regulation mechanism of a previously undescribed photoactivated adenylate cyclase (PAC) from the photosynthetic cyanobacterium Oscillatoria acuminata, showing a central coiled-coil transmits changes from the light-sensing domains to the active sites with minimal structural rearrangement.

3Pos019 マウス微小ウイルスの力学的性質：粗視化分子動力学シミュレーション

Mechanical properties of the minute mice of virus capsid: Coarse-grained Molecular simulation

Koji Ono, Shoji Takada (*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ*)

In virus life cycle, virus regulates its state depending on its environment. Deep understanding of its self-regulatory system is important for synthesis of nano-machines and also for designing new medicine. In this study, we focus on the Minute Mice of Virus (MVM) which is one of the simplest non-enveloped icosahedral single-strand DNA (ssDNA) virus. The MVM formed by 20 equivalent planes, each made of three subunits. To investigate the stability of MVM without ssDNA, we performed coarse-grained molecular simulation (CGMD). We estimated parameters in the protein-protein interactions by comparing B-factor in crystallographic data with the root-mean-square-fluctuation in CGMD. Finally, with these parameters, we analyzed AFM experiments by CGMD simulations.

3Pos020 Momorcharin の酵素活性を制御する活性部位近傍の特殊なコンフォメーション

The specific conformation near the active site regulating the enzymatic activity of momorcharin

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Momorcharin is a Type I RIPs and exhibits *N*-glycosidase activity to inactivating protein synthesis in ribosome. In the present study, the specific conformational changes near the active site induced by the binding of NAG are reported. The FRET distance between Tyr and Trp and segmental rotation of Trp were analyzed by the time-resolved fluorescence spectroscopy. While momorcharin exists in two forms, α- and β-momorcharin, each form showed quite different conformation responding to the binding of NAG. And the enzymatic activity of momorcharin was changed in accordance with the conformational change. Based on the relationship between the conformational change and enzymatic activity, the conformation required for the enzymatic activity of momorcharin was characterized.

3Pos021 ケモカイン受容体の細胞内領域に含まれるケモカインシグナル制御因子フロント結合領域の構造生物学的解析

Structural analyses of the cytosolic region of the chemokine receptor, which interacts with the chemokine signal regulator FROUNT

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The membrane-proximal C-terminal region (Pro-C) is important for the regulation of G-protein coupled receptors, but the Pro-C binding to a cytosolic regulator has not been structurally analyzed. The chemokine receptors CCR2/5 bind to the cytosolic regulator FROUNT. The structures of CCR2 Pro-C bound with FROUNT or biomembrane-mimicking micelles were determined by NMR. Cross-saturation-based experiments revealed that the binding surface on Pro-C for FROUNT overlapped with the binding site for membrane, suggesting competitive binding of Pro-C between FROUNT and membrane. These results support an equilibrium model: chemokine binding changes the conformational equilibrium of CCR2/5 toward the active state, and Pro-C switches its binding partner from the membrane to FROUNT.

3Pos022 酵素と基質と四つのイントロンで形成される平面の関係

Enzyme, Ligand, and Plane formed with Four Introns

Michiko Nosaka, Akari Ichisima, Mami Nakayashiki (*National Institute of Technology, Sasebo College*)

We have identified and evaluated the planes formed with four intron-positions in the tertiary structures of retinol-binding protein and calpain domain-VI. We discussed the significance of the plane in the tertiary structures and evolutional meaning about one of the proteins. (J. Theor. Biol., vol. 340, 139-145, 2014).
Here, we show another examples of enzyme proteins and discuss the relationships between the plane and the ligand in the protein structures.

3Pos023 酵母カーゴ様タンパク質受容体 Emp46p と Emp47p のコイルドコイル領域における pH 依存的な会合と解離

pH-dependent assembly and segregation of the coiled-coil segments of yeast putative cargo receptors Emp46p and Emp47p

Kentaro Ishii[1], Hiroki Enda[2], Masanori Noda[3], Megumi Kajino[2], Akemi Kim[2], Eiji Kurimoto[2,4], Ken Sato[5], Akihiko Nakano[6,7], Yuji Kobayashi[3], Hirokazu Yagi[2], Susumu Uchiyama[1,3], Koichi Kato[1,2,8] ([1]*Okazaki Inst. Integrative Bioscience, NINS*, [2]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [3]*Grad. Sch. Eng., Osaka Univ.*, [4]*Fac. Pharm., Meijo Univ.*, [5]*Grad. Sch. Art. Sci., Univ. Tokyo*, [6]*Grad. Sch. Sci., Univ. Tokyo*, [7]*RIKEN, Cent. Phot.*, [8]*Inst. Mol. Sci., NINS.*)

Emp46p and Emp47p are yeast putative cargo receptors that recycle between the endoplasmic reticulum and Golgi apparatus. These receptors can form complexes in a pH-dependent manner, but their molecular mechanisms remain unclear. Here, we successfully reproduced their interactions *in vitro* solely with their coiled-coil segments, which form stable heterotetramers in the neutral condition but segregate at lower pH. Mutational data identified a key glutamate residue of Emp46p that serves as the pH-sensing switch of their oligomer formation. Our findings elucidate the mechanisms of the dynamic cargo receptor interactions in the secretory pathway and the design framework of the environment-responsive molecular assembly and disassembly systems.

3Pos024 クライオ電子線トモグラフィー法により明らかになったイネ萎縮ウイルスの昆虫細胞からの放出メカニズム

Mechanisms for egress of Rice dwarf virus from insect vector cells revealed by whole cell cryo-electron tomography

Naoyuki Miyazaki[1,2], Akifumi Higashiura[2], Kazuyoshi Murata[1], Atsushi Nakagawa[2], Kenji Iwasaki[2] ([1]*NIPS*, [2]*IPR*)

Newly synthesized viral particles must be released from infected cells for the successful viral proliferation. Here, we examined the virus-release pathway of Rice dwarf virus (RDV), a member of the genus Phytoreovirus in the family Reoviridae, by whole cell cryo-electron tomography (cryo-ET). RDV was inoculated to the host insect vector cells grown on holey carbon supported EM grids for two days. After further cultivation for three to five days, the RDV-infected cells were plunged-frozen in liquid ethane and embedded in vitreous ice for the whole cell cryo-ET. The cryo-ET observations clearly visualized the viral particles releasing from the infected cells, which gave us new insights into the novel mechanisms of the viral egress.

3Pos025 Dynamics of bacterial flagellar filament self-assembly

Anthony Abraham, Takuma Fukumura, Tohru Minamino, Keiichi Namba (*Osaka Uni. Grad. Sch. Frontier Biosciences*)

The bacterial flagellum is a motility organelle. Assembly of the helical filament is the final, continuous step in the morphogenesis of the entire flagellum. The component protein flagellin is exported from the cytosol to the distal end of the growing structure by the flagellar type III protein export system. More than 20,000 flagellin molecules are exported to form the helical propeller. It has been reported that the filament elongation rate decays exponentially with its length. However, a constant elongation rate has also been reported and explained by a chain mechanism. Here, we report the effects of specific deletions in the chain mechanism-dependent regions of flagellin on the export and polymerization of flagellin to discuss the validity of the chain mechanism.

3Pos026 αシヌクレインの添加物によるアミロイド線維形成への影響
The effects additives on the amyloid formation ofα-synuclein

Miki Hirano[1], Masatomo So[1], Hisashi Yagi[3], Yasushi Kawata[2], Yuji Goto[1] ([1]*Institute for Protein Research, Osaka University*, [2]*Dept. of Chem. and Biotech., Grad. Sch. of Eng., Tottori Univ.*, [3]*Center for Research on Green Sustainable Chemistry, Tottori University*)

Although various compounds accelerate or inhibit amyloid fibrillation, some compounds exhibit both effects depending on the concentration. For examples, fibrillation of α-synuclein, associated with Parkinson's disease, was accelerated by low concentrations of SDS, but the concentrations higher than CMC suppressed the fibrillation. Although heparin was reported to accelerate the fibrillation of α-synuclein, we suggested that high concentrations of heparin suppressed the fibrillation. Thus, the concentration-dependent adverse effects might be common to various additives, in which the acceleration was caused by clustering of amyloidogenic proteins through hydrophobic solutes and suppression was caused by the formation of micelle-like complexes or amorphous aggregates.

3Pos027 １アミノ酸置換によるプロリン異性化酵素からタンパク質分解酵素への機能転換
Functional conversion from peptydyl-prolyl isomerase to protease by a single amino acid substitution

Teikichi Ikura, Nobutoshi Ito (*MRI, Tokyo Med. Dent. Univ.*)

Enzymatic function generally consists of three steps, substrate binding, catalytic reaction and product release. The catalytic reaction is often tightly linked to a specific motif like a catalytic triad of serine proteases. Thus, only a few changes of amino acid residues in the motif can change the enzymatic function. A peptidyl-prolyl isomerase, Pin1, catalyzes isomerization of pSer/Thr-Pro bond. Its activity is related to various cellular functions including suppression of Alzheimer's disease. Recently, we find that some mutants of Pin1 showed limited auto-proteolysis. We determined the target sequence by mass spectrometric analysis. We also investigated their proteolysis activities by physicochemical analysis and further mutational approaches.

3Pos028 粗視化力場を用いたタンパク質構造変化を伴うリガンド結合過程の比較解析
A comparative study of the protein-ligand binding processes coupled to protein conformational changes in coarse-grained simulations

Tatsuki Negami, Tohru Terada, Kentaro Shimizu (*Grad. Sch. of Agri. and Life Sci., Univ. of Tokyo*)

Clarifying the mechanism of the conformational change of a protein upon ligand binding is important for understanding its biological function. Molecular simulation is a promising way to provide detailed insight into the ligand binding phenomenon. Previously, we developed a method to enable the conformational change of a protein in a coarse-grained simulation with MARTINI by combining a dual-basin network model with it. Here, we applied this method to two protein-ligand systems and performed five microsecond simulations fifty times with different initial ligand placement around each protein. We found that the ligand binding processes are different between the systems. We will discuss the difference in the ligand-binding mechanism coupled to the conformational change.

3Pos029 脂質による GPCR の活性制御機構の解明
Elucidation of the signal regulation mechanism of GPCRs under physiological lipid bilayer environments

Takuya Mizumura[1], Keita Kondo[1], Takumi Ueda[1,2], Yutaka Kofuku[1], Ichio Shimada[1] ([1]*Grad. Sch. Pha., Univ. Tokyo*, [2]*JST-PRESTO*)

Under physiological conditions, G-protein coupled receptors (GPCRs) are embedded in lipid bilayers that contain docosahexaenoic acids (DHAs) and cholesterols, and some GPCR signalings are regulated by these lipids. However, little is known for their regulation mechanism by the lipid composition of the lipid bilayer, due to the lack of the methods for the investigation of the activities and structures of GPCRs. Here, we embedded adenosine A2A receptors (A2As), one of the GPCRs, into the lipid bilayers in reconstituted high density lipoprotein (rHDL) with various lipid compositions. The G-protein activation by A2A was increased with the increase of the population of DHAs or cholesterols. NMR experiments of A2A in rHDL with various lipid compositions are in progress.

3Pos030 滴定 X 線溶液散乱測定を用いたアダプター蛋白質 GGA-ユビキチン複合体の相互作用/構造解析
Interaction/Structure analysis of GGA-Ubiquitin complex by using titration SAXS Measurement

Miho Shinohara[1], Hironari Kamikubo[1], Keito Yoshida[1], Yoichi Yamazaki[1], Kazuhisa Nakayama[2], Soichi Wakatsuki[3], Mikio Kataoka[1] ([1]*Grad. Sch. Mat. Sci., NAIST*, [2]*Grad. Sch. Pharmaceutical Sciences., Kyoto Univ.*, [3]*School of Medicine, Stanford University*)

GGA involved in a vesicle transport system is a multi-domain protein consisting of VHS, GAT and GAE domains. In a VHS-GAT fragment, VHS and GAT are tightly bound, but VHS is released from GAT (open form) upon binding of MPR to VHS (domain rearrangement). Furthermore, GAT alone can interact with several ubiquitin molecules (Ub). Ub-binding sites of GAT are overlapped with the VHS contact surface. In order to investigate the effect of the domain rearrangements of VHS-GAT on the Ub binding, we carried out Ub-titration SAXS by using our developed auto-sampler. In the result, titration curve for Ub binding is different between the open and the close form of the VHS-GAT fragment, suggesting that the Ub binding is affected by domain rearrangement.

3Pos031 LigMap: 共溶媒分子シミュレーションによるタンパク質のリガンド結合ホットスポットの予測
LigMap: Predicting ligand binding hostspots of proteins by molecular simulations with cosolvent solutions

Hironori Kokubo, Akihiro Yokota, Atsutoshi Okabe (*Takeda Pharmaceutical*)

We have developed a novel computational method, LigMap, which enables us to identify ligand binding hotspots and appropriate drug-like fragment structures by molecular simulations in cosolvent solutions. As an initial test, we applied our method to several publicly-available protein-ligand systems with known ligand binding modes. We found that the positions and orientations of cosolvent molecules predicted by our method were in excellent agreement with the experimental partial structures of ligands obtained from Protein Data Bank. It also identified even hidden pockets which need induced fit of some target proteins properly.

3Pos032 貝由来炭酸脱水酵素ナクレインのカルシウム結合部位の解析
Structural Analysis of Calcium Binding Site in Sea Shell Carbonic Anhydrase, Nacrein

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A shellfish carbonic anhydrase, nacrein, has the Ca^{2+}-binding site (CalB) consisting of the repeat sequence, $(GXN)_{27}$ (X= D, N, Y, or E), and the functional group that catalyzes the hydration reaction of CO_2. A kinetic analysis shows that CalB has an inhibitory function for the formation of $CaCO_3$ crystal. A 298-bp synthetic gene encoding CalB from oyster, *Crassostrea nippona*, was constructed by using PCR. The gene was expressed in bacteria to obtain ^{15}N-labeled CalB. NMR data show that the binding of Ca^{2+} to CalB causes the down-field shift of Asn side chains. Considering that Ca^{2+} interacts with both COOH of Asp/Glu and N atoms of Asn in CalB, there would be the coordination such as $CaCa^{2+}$/EDTA binding. A goal is to develop a catalyst useful for CO_2 capture and storage.

3Pos033 Relative Binding Free Energy Calculation with the Free Energy Variational Principle for fXa-Ligand system

Toshimichi Matsugi, Takeshi Ashida, Takeshi Kikuchi (*Ritsumeikan Univ*)

A computational technique with low cost is highly desirable for the estimation of binding free energies of many molecules. Several techniques have thus far been developed for estimating binding free energies. Some techniques require high large computational cost, and other methods give good predictions but require tuning of some parameters. In this study, we propose a method to predict relative binding free energies with accuracy comparable to the results of prior methods but with lower computational cost and no parameter needing to be carefully tuned. The present technique is based on the free energy variational principle. 2BOH binding protein with 12 ligands is taken as a test system. The details of the results will be presented at the poster session.

3Pos034 高速 AFM による c-Cbl の構造変化の直接観察
Direct observation of conformational change of c-Cbl by high speed AFM

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Ubiquitination is a post-translational modification that regulates cellular functions such as proteasomal degradation, DNA repair, and signal transduction pathway. Ubiquitination of substrate protein occur by transfer of ubiquitin via E1, E2 and E3. It has been reported that ubiquitin transfer activity of c-Cbl is enhanced by phosphorylation, and the structure of phosphorylated c-Cbl is much different from that of none-phosphorylated c-Cbl. It suggests that large conformational changes of c-Cbl during phosphorylation. To investigate the relationship between conformational changes by phosphorylation and ubiquitination, we observed the dynamics of conformational change of c-Cbl by phosphorylation in real time using high speed AFM.

3Pos035 Motion Tree によるタンパク質運動の階層的記述
Motion Tree delineates hierarchical structure of protein dynamics observed in molecular dynamics simulation

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A novel description of protein dynamics is proposed based on the hierarchical clustering of fluctuations in atom-atom distances calculated from molecular dynamics (MD) trajectory. The tree diagram thus constructed, named “Motion Tree”, thoroughly illustrates all sizes, magnitudes, and the cooperativity of dynamic rigid-body domains hierarchically along with the amplitudes of inter-domain fluctuations. We show the applications to adenylate kinase, glutamine binding protein, and a multidrug ATP binding cassette transporter, which have demonstrated the usefulness and remarkable efficiency of the Motion Tree to detect not only collective motions related to large scale structural rearrangements but also local functional motions hidden behind complex MD trajectories.

3Pos036 ATP 結合に伴う GroEL 立体構造変化の計算科学的研究
Computational study of conformational change in GroEL upon ATP binding

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GroEL is a chaperonin that undergoes a conformational change from a closed to an open state. This change requires ATP, but does not require ATP hydrolysis. The following atomic structures are known: closed state with ATP (P state), closed state without ATP (M state) and open state. We conducted the simulations of the conformational change using Elastic Network Model and investigated the effect of ATP binding. We found that the simulation started with P state resulted in reaching a conformation far close to the open state and that that with M state resulted in a conformation further from the open state. It was suggested that an ATP molecule should behave as an insulator that induces population shift from M state to P state that has a pathway leading to the open state.

3Pos037 分子動力学法による好熱性ロドプシンの熱安定性の研究
Assessing Molecular Mechanism of High Thermal Stability of Thermophilic Rhodopsin by Molecular Dynamics Simulation

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Recently, a biochemical and spectroscopic study of thermophilic rhodopsin (TR), derived from an extreme thermophilic bacterium Thermus thermophilus, reported that TR possesses much higher thermal stability than other proton pumping rhodopsins (Tsukamoto et al. Journal of Biological Chemistry, 288, 21581 (2013)). Here, we investigated the high thermal stability of TR at the molecular level by means of molecular dynamics (MD) simulation. We performed two isothermal-isobaric MD simulations at temperatures of 300 K and 348 K under pressure of 1 bar. A slippage of the helix and a simultaneous increase of hydrophobic interactions within TR were observed. This mechanism may account for the thermal stability of TR.

3Pos038 Dissociation and Unfolding of Tobacco Mosaic Virus Coat Protein Assemblages

Hiroaki Fukao[1], Kazumasa Sakurai[2], Yasushige Yonezawa[2], Masao Fujisawa[1], Kazuhiro Ishibashi[3], Masayuki Ishikawa[3], Tetsuo Meshi[3], Hideki Tachibana[1,2] ([1]*Fac Boil-Ortd Sci Tech, Kinki Univ*, [2]*High-Pres Prot Res Center, Kinki Univ*, [3]*Div Plant Sci, NIAS*)

We have produced tobacco mosaic virus coat protein (TMVCP) in E.coli and have shown that at neutral pH denaturant urea of low concentrations dissociates TMVCP disks into oligomers such as trimers, followed by the unfolding of subunit secondary structure with increasing urea concentration. Here we show that the quaternary oligomer state appears to be retained at high urea concentrations where the regular secondary structure should be broken, while at alkaline pH the urea unfolding of the secondary structure takes place in a monomeric state. High propensity of the occurrence of hydrophobic residues in the intrasubunit interfaces between constituent alpha-helices and that of charged residues in the intersubunit interfaces may account for this finding.

3Pos039 NMR で見た水溶液中メリチンの１量体ー4 量体構造転移
Conformational transition between monomer and tetramer of melittin in an aqueous solution studied by NMR

Yoshinori Miura (*Center for Advanced Instrumental Analysis, Kyushu University*)

Melittin, a major component of honeybee venom, is a small polypeptide of 26 amino acid residues. In an aqueous solution it undergoes conformational transition between a random coil monomer and a helical tetramer by variations in temperature, pH and salt content in a solution. The transition correlates closely with the isomers of the proline residue: melittin including a trans proline peptide bond is involved in the transition, whereas melittin having a cis proline peptide bond is not.
We have examined temperature dependence of melittin conformation using NMR spectroscopy in order to clarify thermal stability of the tetramer conformation and cooperativity of the transition.

3Pos040 酸ストレス抗体の中性および穏和な温度における凝集の速度論
Kinetics of antibody aggregation triggered by pH-shift stress at neutral pH and ambient temperatures

Hiroshi Imamura, Shinya Honda (*National Institute of Advanced Industrial Science and Technology*)

Purification process of an antibody involves exposure of the molecules to low pH. The following neutralization causes the aggregation. It remains unclear how the aggregation triggered by pH-shift stress grows at neutral pH and how that depends on temperature in an ambient range. We used dynamic light scattering to monitor time-dependent evolution of size of the pH-shift stressed antibody between 15 and 40 °C. Based on Smoluchowski aggregation kinetics equation, the growth of aggregation triggered by pH-shift stress is described by fractal and reaction limited aggregation. From the temperature dependence, activation energy was determined to be positive, i.e. endothermic, value, which will be key to predict shelf life of the antibody solution.

3Pos041 Role of electrostatic repulsion between unique arginine residues on the assembly of a trimeric autotransporter translocator domain

Eriko Aoki, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. Bioinfo., Grad. Sch. Eng., Soka Univ.*)

Haemophilus influenzae adhesin (Hia) belongs to the trimeric autotransporter family and consists of a passenger domain and a translocator domain. The crystal structure of the translocator domain has shown that this domain forms a 12-stranded transmembrane β-barrel. This protein has a unique arginine residue at 1077. Arg1077 side chains from three subunits protrude from β-strand toward the center of the barrel and close to each other. To investigate the role of this residue on trimer assembly and stability, we replaced this arginine with methionine (R1077M) or lysine (R1077K) and properties of these mutants were investigated. Although the neutralized mutation caused the incorrect oligomer formation, the assembly efficiency of R1077K was similar to wild type.

3Pos042 熱測定を用いたグルカゴン凝集反応の熱力学的研究
Thermodynamic characterization of glucagon aggregation using calorimetry

Tetsuhei Uenoyama[1], Tatsuya Ikenoue[1], Daniel E Otzen[2], Yuji Goto[1], Young-Ho Lee[1] ([1]*Inst. for. Prot. Research, Osaka Univ*, [2]*Department of Molecular Biology, Aarhus Univ*)

We previously characterized thermodynamics of aggregation of several amyloidogenic proteins using isothermal titration calorimetry (ITC). We herein performed ITC-based studies on glucagon aggregation at pH 2.5. Polymorphic properties of glucagon fibrils formed at distinct glucagon concentrations and amorphous aggregation at high NaCl concentrations were distinguished by different enthalpy and entropy changes. A unique thermodynamic parameter such as positive heat capacity changes was also obtained. We demonstrated that ITC was powerful for the thermodynamic study on protein aggregation and thermodynamics of protein aggregation was indeed possible. We propose that thermodynamic properties of amyloid fibrillation are not always consistent with those of protein folding.

3Pos043 Molecular Tailoring Approach 法による蛋白質二次構造内に働く相互作用の量子化学的研究

Quantum chemical study for interactions in protein secondary structures via Molecular Tailoring Approach

Ayumi Kusaka[1,2], Haruki Nakamura[1], Yu Takano[1,3,4] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Grad. Sch. Info. Sci., Hiroshima City Univ.*, [4]*JST-CREST*)

In order to develop a new force field incorporating the characteristics of secondary structures for more accurate molecular simulations for proteins, each hydrogen bond energy in secondary structures was evaluated by Molecular Tailoring Approach with the density functional theory (DFT) at the level of B97D/6-31+G(d). We compared the hydrogen bond energies by DFT to those by the classical force field, FF (AMBER ff99SB), for the typical secondary structures: α-helices, parallel and antiparallel β-structures, modeled with alanine peptides Ace-(Ala)n-Nme. We find that the DFT and FF energies agree well in both parallel and antiparallel β-structures. In contrast, in α-helices, there are remarkable differences, which can be compensated by backbone partial charges.

3Pos044 神経細胞毒性を持つ $A\beta_{42}$ 凝集体と持たない凝集体は共にβ構造から成るが短波長領域で違った CD パターンを持っている。

Neurotoxic and nontoxic $A\beta_{42}$ assemblies are commonly composed of β-sheet structures but have different CD spectra in the short wavelength

Yoshitaka Matsumura[1,2], Tomoya Sasahara[1,2], Takayuki Ohnishi[1,2], Eri Saijo[1,2], Kaori Satomura[1,2], Michio Sato[3], Minako Hoshi[1,2,4] ([1]*TAO Health Life Pharma Co., Ltd.*, [2]*Inst. Biomed. Research & Innov.*, [3]*Meiji Univ.*, [4]*Kyoto Univ.*)

In order to reveal the change in a secondary structure of $A\beta_{42}$ during self-assembly process, we monitored the time-dependent change in CD of $A\beta_{42}$ solutions. Synthtic $A\beta_{42}$ were dissolved in 0.5xPBS 1% acetonitoril/dioctyl phthalate and slowly rotated for 16 hrs at 4℃. In most cases, the CD of the $A\beta_{42}$ solutions showed a unique pattern, which had a small hill between 200-210 nm and a valley around 215-220 nm. In contrast, when we used different synthetic lots of $A\beta_{42}$, their CD showed a typical β-sheet. In spite of this difference, CONTIN indicated that both assemblies lack α-helix but consist of β-sheet. Interestingly, the former $A\beta_{42}$ assemblies showed neurotoxicity against primary rat hippocampal neurons, but the latter have little neurotoxicity.

3Pos045 レプリカ交換分子動力学シミュレーションによる ポリグルタミン酸の pH に対する構造依存性

Structural dependence of poly-glutamic acids on pH studied by replica-exchange molecular dynamics simulations

Ryosuke Iwai[1], Tetsuro Nagai[2], Takuya Takahashi[2] ([1]*Grad. Sci. Life Sci., Ritsumeikan Univ*, [2]*Coll. Life. Sci., Ritsumeikan Univ*)

Structure and function of proteins depend on the solvent pH. In this study, we tried to reproduce pH effects on helix-coil transitions of poly-glutamic acids (PGA), using replica-exchange molecular dynamics (REMD) simulations. We made the model PGA corresponding to an acid condition and a neutrality condition. These models were simulated using four force fields with implicit solvent using Amber. The secondary structure analysis with DSSP and dihedral angles and the analysis of end-to-end distance were performed. These results showed that AMBER-ff03 is effective in reproducing the helix-coil transitions depending on the solvent pH.

3Pos046 枯草菌 F_1-ATPase に於ける DELSEED 領域の機能解析

Role of the DELSEED motif in *Bacillus subtilis* F_1-ATPase

Koji Takada, Yasuyuki Kato-Yamada (*Department of Life Science, Rikkyo University*)

The DELSEED motif, a well-conserved region of β subunit of F_1-ATPase, is thought to have some important roles in torque transmission, and regulation by the ε subunit. Although there are some variations in its primary sequences, the Ser in the center of DELSEED motif is especially well conserved. *Bacillus subtilis* F_1-ATPase (BF_1) is virtually sole example of which has the Gly at this position (DELGEED). We have analyzed the DELSEED mutant of BF_1 to figure out the role of the characteristic DELGEED sequence. It was found that the mutant has different sensitivity to the ε subunit from the wild type. The difference in the conformational change in the ε subunit was also examined.

3Pos047 MD シミュレーションを用いた Neuropsin へのペプチド結合に関する研究

Molecular Dynamics Study of Peptide Binding to Neuropsin

Masami Lintuluoto[1], Mitsumasa Abe[1], Hideki Tamura[2], Yoshifumi Fukunishi[3] ([1]*Grad. Sch. of Life and Environ. Sci., Kyoto Pref. Univ*, [2]*L-StaR, Hoshi Univ. Sch. Pham. and Pham. Sci.*, [3]*AIST*)

Neuropsin is a serine protease expressed locally in the limbic system and is involved in memory formation and psychiatric disorder by its dysfunction. However, the signaling mechanism underlying neuropsin does not fully clarify. The neuregulin-1 (NGR-1) has been known as an important substrate for neuropsin. In this Molecular Dynamics (MD) study, we investigate the interaction between the NGR-1 and neuropsin by using the peptide models for NGR-1. The aim of this work is to evaluate neuropsin's structural and dynamical behavior and relative stability on the neuropsin-ligand complexes, and to obtain new knowledge to reveal the signaling mechanism. We also compare the binding affinity and reaction activity of peptide models to the wild type and C208S mutant of neuropsin.

3Pos048 アクチン結合タンパク質アクチニン-4 とその変異体の機能解析

Predicting the three-dimensional structure of the actin-binding domains of actinin-4 mutants

Nami Miura, Masahiro Kamita, Takanori Kakuya, Hirokazu Shoji, Tesshi Yamada, Kazufumi Honda (*Div. Chem. Clin., Natl. Cancer Ctr. Res. Inst.*)

Actinin-4 (ACTN4), an actin-bundling protein previously identified by our laboratory, is closely associated with cell motility, cancer metastasis, and a variety of cellular processes.
The actin-binding domain (ABD) of ACTN4 consists of a tandem pair of calponin homology (CH) subdomains (CH1 and CH2). The mutants specific to human diseases were restricted to the CH2 subdomain of the ABD; they were located on the side of the helix, facing the helix-helix interface between CH1 and CH2. These findings suggest that the three-dimensional structure of the ABDs of ACTN4 mutants differs from that in wild-type ACTN4, and that these changes are responsible for the biological characteristics of ACTN4-mediated diseases.

3Pos049 MD シミュレーションを用いた G3LEA モデルペプチドとタンパク質の相互作用の解析

Analysis of Group3LEA model peptide-protein interactions by molecular dynamics simulation

Makoto Usui, Takao Furuki, Tadaomi Furuta, Minoru Sakurai (*Center for Biological Resources and Informatics, Tokyo Institute of Technology*)

We here report the results of molecular dynamics simulations for several systems including a model peptide which has two tandem repeats of the characteristic 11-mer motif found in group3 late embryogenesis abundant (G3LEA) proteins and its partner protein (lysozyme and HybD) in aqueous solution. And for comparison, we performed simulations for control peptide which has the same amino acid composition as that of the G3LEA model but scrambled sequence. The G3LEA model was set apart 5 nm from its partner protein in the unit cell. As a result, the G3LEA model approached its partner and formed the association complex. According to detailed analysis, the acidic residues in the G3LEA model contribute to binding with lysozyme and its basic residues do with HybD.

3Pos050 F1-ATPase における ε サブユニットの活性制御因子としての役割

Role of ε subunit as a regulator for F1-ATPase

Makoto Genda[1], Rikiya Watanabe[1], Yasuyuki Yamada[2], Hiroyuki Noji[1] ([1]*Grad. Sch. Eng. , Univ. Tokyo*, [2]*Dept. Sci. , Univ. Rikkyo*)

F1-ATPase is a mechano-chemical energy transducer which synthesizes ATP by a mechanical rotation. This unique energy transduction is regulated by ε subunit of F1, i.e., ε subunit enhances ATP synthesis activity by ~5 folds, however, it remains unclear as to how ε subunit modulates the activity at the resolution of elementary reaction steps. In this study, we conducted a stall-and-release experiment on ε-reconstituted F1 to investigate the impact of ε subunit upon ATP dissociation, an elementary reaction step of ATP synthesis. The ε-reconstituted F1 showed a significant acceleration in ATP dissociation, thereby enhancing ATP synthesis activity. This finding provides the clue for understanding the highly efficient ATP synthesis mechanism of F1 relying on ε subunit.

3Pos051 アデニル酸キナーゼ反応機構に関する計算科学的研究

Computational Study on the Reaction Mechanism of Adenylate Kinase

Kenshu Kamiya (*Dept. Phys., Sch. Sci., Kitasato Univ.*)

Adenylate kinase catalyzes the reaction: ATP + AMP + Mg2+ -> ADP + ADP + Mg2+. We have been studying the theoretical model of the reaction using MM/QM method. We constructed the model of complex structure of enzyme and substrates, ATP and AMP with Mg ion with some water molecules surronding the active center using MM or MD calculation with AMBER99 force field. The truncated models were used for the calculations with ONIOM method, and the reactant, product, transition structures were optimized. The highest level of the theory(B3LYP/6-31+G(d):Amber(embed)) with 853 atoms(89 atoms in QM) gives the reaction barrier of about 19 kcal/mol. The details about the model size, the conformational differences, or the free energy profiles, will be discussed.

3Pos052 高精度で高効率な水和自由エネルギー計算法

An accurate and efficient method to compute the hydration free energy for large and complex molecules

Takashi Yoshidome[1], **Toru Ekimoto**[1], Nobuyuki Matubayasi[2], Yuichi Harano[3], Masahiro Kinoshita[4], Mitsunori Ikeguchi[1] ([1]*Yokohama City Univ.*, [2]*Osaka Univ.*, [3]*Himeji Dokkyo Univ.*, [4]*Kyoto Univ.*)

The hydration free energy (HFE) is essential to understand various chemical processes in solvent. In calculations of the HFE, the explicit treatment of solvent is effective for accuracy, however, it sacrifices computational efficiency. To satisfy both accuracy and efficiency, we decompose the HFE into two parts: one depends on the solute-solvent interaction, and the other is the remaining determined by the geometry of the solute. The former is computed by the all-atom treatment. The latter is computed by a hybrid method of the energy representation method and the morphometric approach, which needs only geometric information. We show the accuracy and efficiency of our method, and demonstrate performance in the discrimination of the native structure from decoys.

3Pos053 分子シミュレーションにおける静電相互作用計算のための新規非エバルト法

Novel non-Ewald methods for calculating electrostatic interactions in molecular simulations

Ikuo Fukuda[1], Narutoshi Kamiya[1], Han Wang[2], Kota Kasahara[1], Haruki Nakamura[1] ([1]*Institute for Protein Research, Osaka University*, [2]*Freie Universitaet Berlin*)

The calculation of the electrostatic interactions requires the most time and needs highly appropriate treatment in classical molecular simulations. To calculate these interactions with high accuracy and low computational cost we have developed the Zero-multipole summation method. The underling physical idea is simple: electrostatically cancelled particle conformations are clipped to define the interactions and essentially use this information via deriving a novel pairwise function instead of the pure Coulombic function. The periodic boundary conditions, which are artificial for most biophysical simulations, are not necessary, and the Fourier part evaluations, which yield complication, are not needed. The details of the theory and numerical results will be presented.

3Pos054 再構築型無細胞タンパク質合成系を用いた 真核生物由来タンパク質の凝集性の大規模解析

Large-scale analysis of aggregation propensities of eukaryotic proteins by using a reconstituted cell-free translation system

Tatsuya Niwa[1], Eri Uemura[1], Kazuhiro Takemoto[2], Shintaro Minami[3], Satoshi Fukuchi[4], Motonori Ota[3], Takuya Ueda[5], Hideki Taguchi[1] ([1]*Grad. Sch. Biosci. and Biotech., Tokyo Tech*, [2]*Dept. Biosci. and Bioinfo., Kyushu Institute of Technology*, [3]*Grad. Sch. Inf. Sci., Nagoya Univ.*, [4]*Faculty Eng., Maebashi IT*, [5]*Grad. Sch. Frontier Sci., Univ. Tokyo*)

To understand the basis of the formation of protein aggregation, we previously conducted comprehensive aggregation analysis of more than 3,000 bacterial proteins under the chaperone-free condition. In this work, we evaluated the aggregation propensities for ~500 cytoplasmic proteins of budding yeast to compare the basis of protein aggregation between prokaryotic and eukaryotic proteome. Unlike bacterial proteins, yeast proteins showed a broader distribution of aggregation propensity. Moreover, the aggregation propensity of yeast proteins correlated with the length of intrinsically disordered regions (IDRs), and the length of IDRs affected the preferences of molecular chaperones. The results suggest that IDRs and chaperones might be connected to protein evolution.

3Pos055 複数の正則化項を用いた圧縮センシングによる NMR スペクトルの再構成

Reconstruction of NMR spectra using compressed sensing with multiple regularization terms

Kazuya Sumikoshi[1], Teppei Ikeya[2], Yutaka Ito[2], Kentaro Shimizu[1] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci. Eng., Tokyo Metropolitan Univ.*)

NMR spectroscopy is used to nondestructively acquire structural information of molecules. Shortening the time required for its measurement will open up new possibilities, e.g., examining proteins in a living cell, where their states change rapidly. One of the approaches to that goal is to reconstruct or infer the actual signal from as few samples as possible. Compressed sensing (CS) is a technique for this sort of inference.
Recently, it has been reported in a few areas, including magnetic resonance imaging, that introducing multiple regularization terms into a framework of CS has improved the reconstruction quality. We therefore evaluated the effect of introducing additional regularization terms other than usual L1-norm into our reconstruction program for NMR data.

3Pos056 アミロイドβ凝集体に結合したチオフラビン T の蛍光寿命

Fluorescence lifetime of Thioflavin T binding to amyloid-beta peptide aggregates

Akinori Oda, Hiroshi Satozono, Tomomi Shinke, Yohei Takata, Hiroyuki Okada (*Hamamatsu Photonics K.K.*)

The fluorescent dye Thioflavin T (ThT) is widely used for detecting and monitoring of amyloid-beta peptide (Aβ) aggregates which are associated with Alzheimer's disease. Nevertheless, the detail of binding modes between ThT and Aβ aggregates is not fully understood. To reveal the ThT-binding sites in Aβ(1-42) aggregates, we measured the fluorescence lifetime. Multi-exponential decay analysis demonstrated that the fluorescence of ThT binding to Aβ(1-42) aggregates can be decomposed into three components with different lifetimes (0.3, 1.3, and 2.5ns). Furthermore, the fluorescence intensities of these components increased depending on aggregation of Aβ(1-42). These results suggest that Aβ(1-42) aggregates have different types of the ThT-binding site.

3Pos057 固体 NMR を用いたユビキチン過剰発現時に単一大腸菌細胞内で合成される分子数の計測

Counting of the molecules synthesized in an ubiquitin-overexpressed intact *Escherichia coli* cell by solid-state NMR

Kazuya Yamada, Ayako Egawa, Toshimichi Fujiwara (*IPR, Osaka Univ.*)

The quantitative analysis of the biological systems in a living cell is essential to understand lives. However there are little methods to obtain the quantitative data. Here, we report a method for counting the number of molecules synthesized during the overexpression of ubiquitin in an intact *Escherichia coli* cell by using quantitative solid-state NMR. The amount of ubiquitin molecules were evaluated from the integral value of the high-resolution ^{13}C NMR of the cells. From these data, the signals of sugar chains such as peptidoglycans were also measured, and the number of ubiquitin and peptidoglycan were evaluated. The number of the cells in a sample were counted by a cell counter. From these results, we calculated the number of the molecules in the cell.

3Pos058 抗酸化亜鉛 VHH 抗体の亜鉛イオン結合特性

Zinc ion binding activity of an anti-ZnO VHH antibody, 4F2

Ryosuke Sasaki[1], Soichiro Kitazawa[2], Ryo Kitahara[2], Yoshikazu Tanaka[3], Izumi Kumagai[4], Mitsuo Umetsu[4], **Koki Makabe**[1] ([1]*Yamagata Univ.*, [2]*Ritsumeikan Univ.*, [3]*Hokkaido Univ.*, [4]*Tohoku Univ.*)

The interaction between biomolecules and inorganic materials has gained much interest for the development of sensors and highly controlled materials in scale and composition. For this purpose, 4F2, a VHH antibody with high affinity and specificity toward zinc oxide (ZnO) surfaces, has been constructed. Here, we report that 4F2 has an affinity toward zinc ion. We determined the binding affinity between 4F2 and Zn ion by using isothermal calorimetry. To elucidate the molecular binding mechanism of 4F2 toward Zn ion, we applied the 2D NMR technique. From observations of chemical shift perturbations, we found that 4F2 recognized Zn ion near the complementarity determining region 3 (CDR3). Our results provide the basis for the recognition mechanism of ZnO by 4F2.

3Pos059 テトラヒメナビオチン化タンパク質の同定と外腕ダイニン運動系への適用

Identification of biotin carboxyl carrier protein in Tetrahymena and its application in in vitro motility systems of outer arm dynein

Masaki Edamatsu (*Grad. Sch. Arts Sci. Univ. Tokyo*)

Axonemal dyneins play central roles in ciliary beating. Recently, a functional expression system of axonemal dynein was established in Tetrahymena. This study identifies biotin carboxyl carrier protein in Tetrahymena (TtBCCP) and demonstrates its application in in vitro motility systems of outer arm dynein. The TtBCCP is composed of about 70 amino acids and located at the C-terminal of carbamoyl phosphate synthase-like protein. The TtBCCP does not contain the thumb loop and was efficiently biotinylated in Tetrahymena. The TtBCCP-fused outer arm dynein glided microtubules with a velocity similar to that of native dynein, and moved along microtubules in the TIRF assay. The TtBCCP tag is useful for molecular studies of axonemal dyneins and ciliary beating.

3Pos060 細胞透過性コイルドコイルタンパク質の熱安定性

Thermal stability of coiled-coil cell-penetrating proteins

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Molecules with structural anisotropy and rigidity, such as asbestos, demonstrate high cell-penetrating activity but also high toxicity because of their non-biodegradability. Previous study, we synthesized the artificial cationic coiled-coil protein, CCPC 140, was designed to have the structural frame of human skeletal muscle alpha-tropomyosin. This CCPC140 showed superior cell-penetrating activity. Looking at thermal melting profiles of circular dichroism, although all the amino acid residues at the coiled-coil intramolecular interface in heptad repeat were conserved, the Tm of CCPC 140 was much higher than that of alpha-tropomyosin. This indicates that the residues at outer surface of coiled-coil motif can be a determinant of coiled-coil structure stability.

3Pos061 Selection of RuBpy3 motifes from a randomized peptide library

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Unique redox properties of Tris(bipyridine) ruthenium(II) have contributed to diversify its applications in recent years. The introduction of these properties into redox proteins will provide effective means for potential applications in a wide variety of areas. Thus motivated we aim to discover Ru-binding motifs by using genetically encoded unnatural amino acids; an archaeal set of an amber suppressor tRNA (CUA) and a mutant of TyrRS incorporates bipyridylalanine (BpyAla) at amber stop codon without crosstalk with the E.coli translation system. So far we confirmed the incorporation of 3BpyAlas into a peptide sequence by MS. The incorporation of three BpyAlas into randomized sequences is currently under investigation.

3Pos062 ヒト cytochromeb561 ホモログタンパク質・ヒト SDR2 の生理機能解析

Functional analysis of human stromal cell-derived receptor 2, a homolog of cytochrome b561

Rei Toda, Yuma Takahashi, Takako Yamazoe, Akikazu Asada, Motonari Tsubaki (*Dept. of Chem., Grad. Sch. of Sci., Kobe Univ.*)

Cytochrome b561 is a membrane protein, having a role for regeneration of vesicular ascorbate by transmembrane electron transfer. Stromal cell-derived receptor 2 (SDR2) is a homolog of cytochromeb561 and is characterized by the presence of two hydrophilic domains, Reelin and DOMON fused in tandem at the N-terminus of cytochrome domain. Reelin domain is homologous to N-terminal region of Reelin protein that regulates processes of neuronal migration and positioning in developing brain. DOMON domain, is found in N-terminal region of DOpamine beta-MOnooxygenase. Physiological function of SDR2 and roles of the two domains are not known. We investigated intercellular localization of SDR2 in cultured HeLa cell and found that SDR2 is expressed in endoplasmic reticulum.

3Pos063 QM/MM と MD シミュレーションを用いた黄色ブドウ球菌の細胞壁上の高速ヘム輸送機構の解明

Rapid heme-transfer reactions across the cell wall of *Staphylococcus aureus*: a theoretical study using QM/MM and MD simulations

Yoshitaka Moriwaki, Tohru Terada, Kentaro Shimizu (*Dept. Biotech., Grad. Sch. Agri., Univ. of Tokyo*)

IsdH-N3, IsdA-N, and IsdC-N domains of *Staphylococcus aureus*, which are located in its cell wall, transfer heme toward the cell membrane and share similar tertiary structures (RMSD < 2 A). The mechanism of the rapid transfer of heme between these proteins has remained unclear. Here, we propose models of the heme-transfer intermediate complexes. In this model, heme is sandwiched between the donor and acceptor Isd domains. We found that the model structures were stable during 500-ns MD simulations. Subsequent QM/MM (ONIOM) structural optimization showed that the tyrosine residues of the two Isd domains were simultaneously coordinated to the iron atom of heme. Thus, the Isd proteins efficiently transfer heme while forming a pseudo-symmetric protein-protein complex.

3Pos064 共鳴ラマン分光法による 2 価コバラミンの pH に依存した構造変化の検出

Resonance Raman Detection of Cobalamin (II) : pH-Dependent Structural Change

Kaoru Mieda, Takashi Ogura (*Grad. Sch. Sci., Univ. Hyogo*)

Vitamin B_{12} serves as the cofactor(Cbl) in several enzymes that play physiologically important roles in metabolism. It is involved in methyl group transfer reaction in methylmalonyl-CoA mutase. It was demonstrated that pH-dependent spectral changes of Cbl(II) took place [1]. The presence of six- and five-coordinated Co was suggested for the alkaline and acidic sides of the pKa=4.8. Vitamin B_{12} deficiency causes wide range of symptoms and has been studied extensively. In this study, we measured the resonance Raman spectra of Cbl(II) in the presence of SO_2^- at pH 8 and pH 3. In the spectra, we detected the changes of Raman bands due to pH dependent structural changes.

Ref. 1. Salnikov et al. (2011) Dalton Trans. 40, 9831-9834.

3Pos065 呼吸鎖 O_2 還元酵素反応中間体の構造解析

Structural analysis of respiratory O_2 reductase in the reaction intermediate state

Kazumasa Muramoto (*Dept. of Life Sci., Univ. of Hyogo*)

Respiratory chain generates proton motive force coupled with electron transfer at high energy efficiency. O_2 and NO molecules are used as terminal electron acceptor. Respiratory O_2 reductases (O_2Rs) and NO reductases (NORs) belong to evolutionally related superfamily. O_2Rs are broadly classified into A, B and C-types based on their molecular structures.

In previous study, we reported x-ray structure of bovine A-type O_2R in the reoxidized state (Fujisawa *et al.*, 49th meeting 2011). In this study, I further analyzed structure of the O_2 reduction site and compared it with other enzymes in the superfamily. Two water molecules were newly found nearby the site. Two ligand oxygen atoms in the site are close together within a distance shorter than typical hydrogen bond length.

3Pos066 光照射固体 NMR による 13-cis, 15-syn フォトサイクル及び中間体の解明

Elucidation of 13-cis, 15-syn photocycle and its intermediate by photo-irradiation ss-NMR

Arisu Shigeta[1], Kyosuke Oshima[1], Izuru Kawamura[1], Takashi Okitsu[2], Akimori Wada[2], Satoru Tuzi[3], Akira Naito[1] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Kobe Pharm. Univ.*, [3]*Univ. Hyogo*)

Bactriorhodopsin (BR) is a one of the well-studied membrane protein which functions as a light-driven proton pump. BR has two retinal configuration of all-trans (AT) and 13-cis, 15-syn (CS) with 1:1 ratio in the dark state, and changes to ~100% AT under photoirradiation at 20℃. By means of photoirradiation ss-NMR, CS-like intermediate was trapped during the pathway from dark-adapted state to light-adapted state at under -20℃. As was suggested to be a one of intermediates in a CS photocycle which has no proton pumping function, this may reveal the reason why BR has not only active AT retinal but also non-active CS isomerization. Structure of CS-like intermediate will be discussed at poster presentation comparing to N intermediate, active intermediates in AT photocycle.

3Pos067 G 蛋白質共役受容体を熱安定化させるアミノ酸置換の予測：自由エネルギー関数の開発

Prediction of Thermostabilizing Mutations for G Protein-Coupled Receptors: Development of Free-energy Function

Satoshi Yasuda[1], Yuta Kajiwara[2], Yuuki Takamuku[3], Nanao Suzuki[3], Takeshi Murata[3], Masahiro Kinoshita[1] ([1]*Institute of Advanced Energy, Kyoto Univ.*, [2]*Graduate School of Energy Science, Kyoto Univ.*, [3]*Graduate School of Science, Chiba Univ.*)

Structural determination of G protein-coupled receptors (GPCRs) has been hindered by their low stability in detergents. Introducing mutations into GPCRs can enhance their stability, and the random search is currently employed to find those leading to stability enhancement. However, it suffers very low success rate and requires heavy experimental burden. Here we develop a free-energy function (FEF), which is focused on the translational entropy of nonpolar chains of the lipid bilayer and on the hydrogen bonding, for constructing a powerful method of predicting thermostabilizing mutations. As the first step, we test our FEF for the adenosine A2a receptor whose wild-type structure is known and utilized. We demonstrate its high success rate with the aid of experiments.

3Pos068 アクアポリン 1 の細孔口における水分子のダイナミクス

Water dynamics at channel entrance of aquaporin 1

Eiji Yamamoto[1], Takuma Akimoto[1], Masato Yasui[2], Kenji Yasuoka[3] ([1]*Department of Mechanical Engineering, Keio University*, [2]*School of Medicine, Keio University*, [3]*Department of Mechanical Engineering, Keio University*)

Aquaporins (AQPs) transport water molecules across cell membranes selectively. The water pathways in AQP molecules have a characteristic hourglass shape. Water molecules are transported in the central channel in single file and exhibit a non-Poisson feature, which is attributed to the 1/f fluctuations of amino acids [E. Yamamoto et al., Phys. Rev. E 89, 022718 (2014)]. Water pathways connect to bulk water reservoirs via a conical entrance. However, the behavior of water at these channel entrances remains unclear. In this work, using all-atom molecular dynamics simulations, we analyze water dynamics at the conical entrance and study the interactions between amino acids and water molecules.

3Pos069 FSEC を用いた膜タンパク質複合体 MotA/B の構造安定性評価

Evaluation of structural stability of membrane protein complex MotA/B by FSEC

Hiroko Takazaki[1], Keiichi Namba[1,2], Takashi Fujii[1,3] ([1]*Grad. Sch. of Frontier Biosci., Osaka Univ.*, [2]*QBiC RIKEN*, [3]*JST PRESTO*)

MotA and MotB form a proton channel complex (MotA/B) that functions as the stator of proton-driven bacterial flagellar motor. The structure of this complex is essential for understanding motor mechanism but still remains elusive. Toward elucidation of the MotA/B structure, we must find a highly stable MotA/B complex. FSEC (fluorescence-detection size-exclusion chromatography) is a powerful tool for high-throughput pre-crystallization screening of membrane proteins and requires fusion of target proteins with fluorescent proteins. Now, we are constructing GFP-fused MotA or MotB from several thermophilic bacterial species at the N- or C-terminus to investigate expression and structural stability of the MotA/B complexes for structural analysis.

3Pos070 Toward simultaneous observation of conformational dynamics and proton transport in FoF1-ATP synthase

Naoki Soga[1], Rikiya Watanabe[1,2,3], Mayu Hara[1], Hiroyuki Noji[1,3] ([1]*Dept. of app. chem., The Univ. of Tokyo*, [2]*PRESTO, JST*, [3]*CREST, JST*)

FoF1 ATP synthase (FoF1) is the rotary motor protein, which couples mechanical rotation to proton translocation across bio-membrane. To elucidate the unique coupling mechanism, the simultaneous observation of the rotation with proton translocation of FoF1 has been awaited with great interest. Recently, we developed an arrayed lipid bilayer chamber system (ALBiC), which allows to measure the proton translocation of FoF1 at single molecule level. In this study, we conducted the rotation assay of FoF1 on ALBiC for the simultaneous observation. The results showed that some molecules rotate in anticlockwise driven by ATP hydrolysis, which presumably couples to proton translocation. We will report the analyzed data and discuss the prospective of the simultaneous observation.

3Pos071 パッチクランプ法による大腸菌呼吸鎖の末端酸化酵素の H^+-pump の機能解析

Patch clamp studies on the terminal oxidase of *E. coli*

Masaaki Uno[1], Kosuke Komazawa[1], Teruo Kuroda[2], Hisashi Kawasaki[1], Isamu Yabe[1] ([1]*Grad. School of Adv. Sci. and Tech., Mate. and Life Sci., Tokyo Denki Univ.*, [2]*Grad. School of Med., Dent and Phar Sci., Okayama Univ.*)

Patch clamp (PC) is an excellent method for evaluating the function of the transport system. The PC method can be used to directly measure the activity of the ion transport system present on the cell membrane and allows for control of the driving force from the outside. In this presentation, we measured the ubiquinol-induced current of Cyt *bo* alone by applying the PC method to the Cyt *bd*I Cyt *bd*II double gene-disrupted strain and the Cyt *bd*I Cyt *bd*II ClCa ClCb quadruple gene-disrupted strain using 135 mV reversal potential of the V-I curve and $\Delta pH = 2.5$ ($pH_{in} = 7.4$, $pH_{out} = 4.9$), which was loaded both in and out of the membrane. We evaluated the 285 mV as a proton-motive force in Cyt *bo*.

3Pos072 標的分子に適したナノポアの再構成と一分子検出への挑戦

Reconstitution of a target-matching nanopore and challenge for a single molecule analysis

Hirokazu Watanabe, Ryuji Kawano (*Dep. of Biotec. and Life Sci., Tokyo Univ. of Agr. and Tech.*)

Nanopore sensing is a strong tool for a single molecule analysis of the translocating molecules through a nanopore. This method has studied as a highly sensitive biochemical measurement at a single molecule level, such as DNA sequencing and determining the molecular weight of water-soluble polymers. The larger-sized molecules than pore diameter are not able to translocate the pore, and these molecules cannot be detected. Although a greater variety of nanopores are required for the sensing of extensive targets, only a few types of nanopore have been used. In this study, we reconstituted several nanopores using membrane proteins or peptides. We consider that these nanopores are available for food inspection or disease diagnosis by analyzing toxic or marker proteins.

3Pos073 カロテノイド結合に伴うハロロドプシンの熱安定化
Thermal stabilization of halorhodopsin by binding of carotenoid

Takanori Sasaki[1], Kaede Suzuki[1], Takashi Kikukawa[2], Makoto Demura[2] ([1]*Sch. Sci. and Tech., Meiji Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*)

A light driven anion pump halorhodopsin (NpHR), which exists on the membrane of *N. Pharaonis*, forms trimer and binds a carotenoid of bacterioruberin (BR). Trimer NpHR obtained from *E.coli* over-expression system also can bind the BR in the detergent system.
In this study, we examined the effect of the BR binding for thermal stability of the NpHR in the pH range of 7.0-8.5. By thermal treatment at 40 ℃ for 60 min, bleaching ratio of NpHR unbound BR was about 52 % at pH 8.5, 14 % higher than that at pH 7.0. Contrary to this, bleaching ratio of the NpHR-BR complex was about 20 % at pH 8.5, and only 6 % higher than that at pH 7.0. These results suggest that the BR binding contributes to the stabilization of the NpHR not only at neutral pH but also at alkali condition.

3Pos074 高速原子間力顕微鏡によるバクテリオロドプシン球殻構造体の分子構造の観察
Molecular arrangement in bacteriorhodopsin vesicles observed by high-speed atomic force microscopy

Yuto Noda, Daisuke Yamamoto (*Grad. Sch. Sci., Fukuoka Univ.*)

Bacteriorhodopsin (bR) is a light-driven proton pump that forms two-dimensional crystal, called purple membrane (PM). In the presence of a detergent, PM spontaneously converts its shape from a flat membrane into a uniformly-sized vesicle. So far, the molecular arrangement in the bR-vesicle is uncertain. Here, we applied high-speed atomic force microscopy to directly observe the intramembrane structure of the bR-vesicle. The bR-vesicle was observed as a flat membrane on mica surface. In the presence of a low concentration of the detergent, bR molecules showed regular arrangements in the membrane. On the other hand, in the presence of a high concentration of the detergent, bR molecules showed heterogeneous arrangements and dynamically diffused in the membrane.

3Pos075 分子シミュレーションによる転写因子の DNA 上障害物の迂回機構の研究
How transcription factor bypasses obstacles bound on DNA studied by molecular simulations

Mami Saito[1], Tuyoshi Terakawea[2], Shoji Takada[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*Univ. Columbia*)

According to single molecular experiments, transcription factors search their target sites with sliding on DNA and 3D diffusion. These experiments mainly were under the conditions of liner DNA without obstacles in vitro or few information about the positional relation between obstacles and DNA in vivo. But the positional relation of obstacles such as nucleosome or other transcription factors and DNA should have a strong effect on sliding and 3D diffusion. So we tried to reveal the searching mechanism with coarse-grained molecular dynamics software, Cafemol.

3Pos076 H3 ヒストンテールのアセチル化はその立体構造にどのような影響を与えるか
How does an acetylation affect the conformation of H3 histone tail?

Jinzen Ikebe[1], Shun Sakuraba[2], Hidetoshi Kono[1] ([1]*MMS, JAEA*, [2]*GSFS, U Tokyo*)

In eukaryotic cells, genome DNA is stored in a complex with histone proteins (H3, H4, H2A and H2B). Acetylation to terminal regions of histones (histone tails) is generally believed to regulate gene expression through dissociation of tails from DNA, although conformations of disordered tails remain poorly understood. In this work, we examined differences in conformational ensembles of H3 tail with or without K14 acetylation using adaptive lambda square dynamics simulation. The result suggested that the acetylation does not make the tail dissociate from DNA. Instead, it enhanced secondary structure formation of the tail and unwrapping of DNA from the structured histone core regions. This study elucidated the first step of the gene regulation mechanism.

3Pos077 バイモーダルな mRNA コピー数分布の分子起源
Molecular origins of bimodal mRNA copy-number distribution

Keisuke Fujita[1], Mitsuhiro Iwaki[1,2], Toshio Yanagida[1,2] ([1]*QBiC, Riken*, [2]*Grad. Sch. of Front. Biosci., Osaka Univ.*)

In this study, we reconstructed prokaryote transcription in vitro system and visualized the transcription process by using fast fluorescence in situ hybridization (fastFISH). Our results show that in our in vitro system the distribution of mRNA copy numbers follows a bimodal distribution. Based on previous reports, we hypothesized this bimodal distribution was the result of pauses in the transcription elongation of a short DNA template. Furthermore, we found that this bimodality depends on RNAP concentration. From these results, we propose a molecular model where binding of a second RNAP on a template shortens the pause time of the original RNAP by literally pushing it off, contributing to the bimodal distribution of mRNA copy numbers and stochastic gene expression.

3Pos078 親水性ポリマーがある混雑効果によって引き起こされた DNA の折り畳み転移: PEG の分子サイズの効果
Folding transition of DNA induced by crowding effect with hydrophilic polymers: Effect of molecular size of PEG

Shogo Ogata[1], Kenichi Yoshikawa[1], Takahiro Kenmotsu[1], Yuko Yoshikawa[2] ([1]*Doshisha University*, [2]*Ritsumeikan University*)

Living cells maintain their lives in a highly crowding condition. We have studies the crowding effect on the higher-order structure of DNA with PEG of different molecular weight, by use of single DNA observation. It was found that the weight concentration to induce the folding transition, from coil-to-globule state, on DNA becomes smaller with the increase of the molecular weight, or degree of polymerization, of PEG. We will also report the result of our observation on the effect of salt and some other physic-chemical parameters on the folding transition. The mechanism of the folding transition caused in crowding conditions with polymer will be discussed in terms of depletion effect.

3Pos079 Sequence dependence of the stability of single-strand base-stacking

Ryosuke Murai[1], Hiroaki Hata[2], Akira Suyama[1] ([1]*Grad. Sch. Arts and Sci., Univ. Tokyo*, [2]*Grad. Sch. Sci., Univ. Hyogo*)

The single-strand base-stacking (SSBS) is known to affect interactions between nucleic acid strands, and thus the SSBS has a considerable influence on biological functions of nucleic acids as well as technologies using nucleic acids. However, the sequence dependence of the stability of SSBS has not been characterized quantitatively. In this study, we conducted classical molecular dynamics simulations on DNA base quadruplets to investigate the sequence dependence of SSBS stability. The results showed that the stability varied widely from sequence to sequence and had no fully positive correlation with the content of purine bases known to form a stable SSBS. We will systematically discuss about the mechanism causing the sequence dependence of the stability of SSBS.

3Pos080 Promotion & Inhibition of Gene-Expression with Polyamines

Ai Kanemura[1], Yuta Shimizu[1], Yuko Yoshikawa[2], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Doshisha University*, [2]*Ritsumeikan University*)

We have studied the effect of polyamines on the efficiency of gene expression by adapting a cell-free expression system in vitro. It is found that polyamines accelerate the reaction of gene expression at their low concentration. Whereas, they inhibit the expression completely at high concentrations. When we examine sprmine(4+), maximum promotion was observed around 0.05mM, and almost perfect inhibition was caused above 0.3mM.
We will show the difference on the profile of such kind of bimodal effect of polyamines, depending on their structural specificity of the chemical structures. Especially, the bimodality will be argued in relation to the intrinsic character of DNA to generate on/off type of folding transition.

3Pos081 Protect Effects of Ascorbic Acid against Double-strand Breaks in Giant DNA Molecules: Comparison among the Damages

Yue Ma[1], Naoki Ogawa[1], Yuko Yoshikawa[2], Toshiaki Mori[3], Tadayuki Imanaka[2], Kenichi Yoshikawa[1] ([1]*Doshisha University*, [2]*Ritsumeikan University*, [3]*Osaka Prefecture University*)

The protective effects of ascorbic acid on DNA's double-strand breaks were studied by single-molecule observation through fluorescence microscopy. Three different kinds of radiation sources, visible light, γ-ray, and ultrasound, which caused the double-strand breaks, had been used in the experiment. Through quantitative analysis of the experimental result, it was found that ascorbic acid effectively protects DNA molecules against the irradiations by visible light and γ-ray, whereas ascorbic acid shows no apparent protective effect against ultrasound. We will discuss the difference on the protective effect of ascorbic acid in relation to the physic-chemical mechanisms of double strand breaks with different radiation sources.

3Pos082 複製開始タンパク質 DnaA と二本鎖 DNA 相互作用の分子動力学計算による解析
Interaction of replication initiator protein DnaA with dsDNA studied by molecular dynamics simulation

Masahiro Shimizu, Shoji Takada (*Grad. Sch. Sci., Kyoto Univ.*)

Bacterial replication initiator protein DnaA form replication initiation complex on the replication origin *oriC* . This complex unwinds dsDNA and recruits DnaB helicase onto the unwound *oriC*. DnaA can also regulate their activity by binding specific genome region with DNA bending proteins. Some of DnaA sites are bent by DnaA, and this bending is important for forming functional complex. But, sequence dependency of DnaA-dsDNA complex structure is unknown. In this study we prepared 22 *E. coli* DnaA domain IV-dsDNA complex structures that varied in base sequence and conducted systematic full-atomistic molecular dynamics simulations. We report the base sequence dependency of the structure of *E. coli* DnaA domain IV-dsDNA complexes.

3Pos083 SEVENS 法：ヌクレオソームの局所密度の違いでクロマチンを分画する
SEVENS assay: a chromatin fractionation based on the local density of nucleosomes

Satoru Ishihara (*Fujita Health Univ. Sch. Med.*)

Gene expression is correlated with the magnitude of genome packaging; active genes are encoded in open chromatin, while repressed genes in closed chromatin. Most of common methods to access chromatin are specialized for the openness of chromatin, but a non-open structure could not be judged as closed chromatin. Together with recent reports that chromatin is dynamically changed between an open and a closed states, such a structure should be evaluated as graded structure. Here, we utilized a short crosslinker, formaldehyde, capable of crosslinking between neighboring nucleosomes. Using a sedimentation velocity centrifugation, chromatin treated with this reagent was sequentially fractionated in an aspect of the local density of nucleosomes.

3Pos084 酵素を用いた DNA 反応系によりつくられた XOR 演算回路
XOR circuit constructed using enzymatic DNA reactions

Toshihiro Kojima, Yoko Sakai, Koh-ichiroh Shohda, Akira Suyama (*Univ. of Tokyo*)

RTRACS developed by our group is a molecular computer that processes information using an enzymatic DNA circuit. The circuit is made from assemblies of basic modules that are capable of performing various operations including logical operations. In this study, we experimentally constructed an XOR (exclusive OR) circuit using three logic gate modules (LGMs). The LGM is one of basic modules of RTRACS and can perform multiple operations, including AND, OR, NAND and NOR. The XOR circuit constructed was closely examined using molecular beacon probes and gel electrophoresis. The results demonstrated that the circuit performed the operation almost as designed. We will also discuss about the potential of RTRACS to construct larger DNA circuits.

3Pos085 分子シミュレーションを用いた２成分溶液系における選択的溶媒和の部分モルエンタルピーへの影響の研究

Effects of Preferential Solvation on Partial Molar Enthalpies in Binary Mixture Systems: Molecular Simulation Study

Yuichi Kawabata, Ryo Akiyama (*Grad. Sch. Sci., Univ. Kyushu*)

Excess partial molar enthalpies in binary mixtures are often said to represent the changes of interactions between the components, although microscopic understandings are not clear. In this study, we expressed the partial molar enthalpy of the solute in binary mixture with three terms (direct interaction, solvent reorganization, and non-local term) based on the solvation shell model, and evaluated each contribution for simple systems using molecular simulation. We found the non-local term which reflects the composition changes in bulk space arising from preferential solvation appears if the solvation process is isobaric-isotherm. In the presentation, we'll show the new expressions with derivation and discuss the interpretation of simulation results.

3Pos086 The first application of adaptive quantum mechanics / molecular mechanics method for infrared spectra of liquid phase

Hiroshi Watanabe[1], Misa Banno[1], Kubar Tomas[2], Elstner Marcus[2], Minoru Sakurai[1] ([1]*Tokyo Inst. of Tech.*, [2]*Karlsruhe Inst. of Tech.*)

Quantum effect in solute-solvent interaction such as many-body effect and dipole-induced dipole is known to be critical factor for infrared spectrum in liquid phase. Thus, in order for accurate spectrum evaluation, not only a solute of interest but also surrounding solvent molecules should be treated with quantum mechanics method. However conventional quantum mechanics / molecular mechanics (QM/MM) simulation cannot handle free quantum mechanics (QM) solvent molecules in molecular dynamics simulation because of diffusion problem. To deal with this problem, we proposed an adaptive QM/MM, "Size-Consistent Multipartitioning method" and as the first application demonstrates reproduction of infrared spectrum in aqueous system.

3Pos087 3 次元分布相関を基にしたタンパク質水和構造予測法の開発

Development of structural sampling for predicting hydration structure around the protein, based on 3-dimensional distribution function

Shunsuke Chiba, **Yasuomi Kiyota**, Mayuko Takeda-Shitaka (*Kitasato University*)

In protein function analysis or drug design studies, hydration structure around the protein is important to understand the binding mechanism. For predicting the hydration structure, there are many theoretical methods like MD simulation, but these methods have complex processes, because of higher entropy of water. Therefore, we developed a structural sampling method based on the 3D-distribution function with Inversed-transformed Monte-Carlo. The 3D-RISM theory can calculate the hydration structure as a 3D-distribution function, directly. The distribution function is difficult to image the molecularly hydration structure. This method can reproduce the conformation of hydration structure. We applied this method to some proteins which have notable hydration structure.

3Pos088 溶質分子周囲の水分子ダイナミクスの MD シミュレーション：人工的に導入された LJ ポテンシャルパラメタ変化の影響

MD simulations of water dynamics around solute molecules: Effect of LJ potential parameter changes artificially introduced

Takuya Takahashi, Tetsuro Nagai (*Coll. Life. Sci., Ritsumeikan Univ.*)

Many experiments and calculations suggest that mobility of water molecules around solute molecules changes according to the solute nature. For example, water molecules around structure-breaker solutes have larger translational self-diffusion coefficients than a water molecule in bulk has. Here, we tested a set of suitable Lennard-Jones potential parameters (LJ) of interactions between modified TIP5P water model and solutes to investigate the dynamical behavior of water molecules. In addition, we analyze the number of hydrogen bonds and the radial distribution functions with a number of combinations of LJ values, with increased space resolution. We then investigate their correlations to the dynamical behavior to study how the water dynamical behavior is governed.

3Pos089 Wnt/β-catenin と FGF/ERK シグナルは再生皮膚の羽毛原基の発生とパターニングに関与する

Wnt/β-catenin and FGF/ERK signaling are involved in the feather bud formation and patterning of reconstructed skin

Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

Wnt/β-catenin and fibroblast growth factor (FGF)/ERK signaling are known as key pathways in the development of ectodermal organs such as feather, hair and tooth. We have developed a bioengineered chick skin and feather buds that were self-organizing the size and spacing of native embryonic development through epithelial-mesenchymal interactions. Inhibition of FGF signaling by treating with inhibitors led to fusion of the feather buds and up-regulated wnt-related genes including CTNNB1 and LEF1. Furthermore, activation of Wnt signaling by GSK-3β inhibitor resulted in the expansion or fusion of the feather bud regions. These results suggest that Wnt/β-catenin and FGF/ERK signaling are cooperatively involved in the bioengineered feather bud formation and patterning.

3Pos090 Mapping elastic modulus of mouse submandibular gland tissue by atomic force microscopy : effect of cytoskeletal modification

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Branching morphogenesis is a developmental process appearing in various organs. Submandibular gland tissue (SMG) has been widely used as a model organ to investigate the mechanism of branching morphogenesis. Recent studies indicate that mechanical properties such as traction force and elasticity of cells play an important role on the development of SMG[1], but the detailed relationship has not been elucidated.. In this study, we measured the topography and the elastic modules of mouse SMG by atomic force microscopy. We found that those values were highly correlated, and the correlation was reduced as the actin filaments were depolymerized, implying that the actin filaments mechanically regulate the branching morphogenesis.

[1] Miyajima et al. Biomaterials 32(2011)6754.

3Pos091 C. elegans 受精卵の極性維持には RING タンパク PAR-2 の膜-細胞質間の local な交換反応が機能する

Cortical polarity gradient maintenance by local-rapid cortex-cytoplasm exchange of a posterior PAR

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Conserved partition-defective (PAR) polarity proteins are thought to diffuse across the anterior-posterior (a-p) boundary during the polarity maintenance in C. elegans embryos. Contrastingly, we identified novel cortical PAR-2 components that dissociated in too short times to diffuse across the a-p boundary using single-molecule imaging. A mathematical model based on the quantitative measurements reproduced the asymmetric PAR-2 localization. We conclude that local-rapid exchange throughout the cortex mediated by the "short-residing" components is a fundamental mechanism for polarity maintenance in C. elegans embryos. The novel components explain a biphasic distribution of polarity proteins along the polarity axis in C. elegans embryos and other polarized cells.

3Pos092 横紋筋筋原線維束上の２次元 SPOC 波に関するモデルシミュレーション

Model simulation on the two dimensional SPOC wave in a bundle of striated myofibrils

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SPOC (Spontaneous Oscillatory Contraction) is the phenomenon observed in striated muscle under the intermediate activation conditions. Recently, we constructed the theoretical model of SPOC for a sarcomere, a unit model, which explains the behavior of SPOC (Sato, K. et al., 2011). We also constructed a connected model, visco-elastically connecting the unit models in series, which explains the behaviors of SPOC at a myofibril level (Sato, K. et al., 2013). This model could reproduce the propagation patterns of SPOC wave, and determin critical parameters that define various SPOC patterns. In the present study, we extended the connected model to a 2D model, in which myofibrils were elastically connected in parallel (side-by-side), and examined 2D patterns of SPOC wave.

3Pos093 水晶振動子上でのアクトミオシンの質量

Weighing the apparent mass of actomyosin by QCM

Kazuya Soda[1], Takashi Ishiguro[2], Hajime Honda[1] ([1]*Dept. of Bioeng., Nagaoka Univ. Tech.*, [2]*Taiyo Yuden Co., Ltd.*)

In recent years, solvent molecules were considered to be involved in the interactions between actin and myosin both in vivo and in vitro. In order to elucidate the role of solvents, we have measured the apparent mass of actin filaments interacting in vitro with HMM molecules spread along the surface of QCM (Quartz Crystal Microbalance) electrode. Decreases of frequency upon attaching of actin filaments and increases upon addition of ATP were successfully measured. However magnitude of the frequency change was found to be much larger than that calculated from the molecular mass of actin filament. The results insinuate that the solvent molecules were cooperatively involved in the interaction between actin and myosin.

3Pos094 ミオシンサブフラグメント 1 の水和に及ぼす ATP アナログの影響

Hydration analysis on myosin subfragment-1 with ATP-analogs

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Myosin is a motor protein which drives an actin filament using the ATP hydrolysis energy. To understand the energetics and motor mechanism, changes in the structure and hydration states of myosin subfragment-1 (S1) intermediates during the chemo-mechanical cycle must be resolved. In this study, dielectric relaxation spectroscopy (DRS) was used to analyze the hydration states of the myosin ATPase-intermediate analogs using AMPPNP, ADP.BeFx, ADP.AlF4, and ADP.Vi, because the real intermediates were difficult to be measured directly. As a result, the restrained water of S1 was increased by binding with those ATP-analogs. This result opposed to the result of the previous transient-measurement study made by one of the authors (Suzuki et al., Biophys J 1997,72, 18-23).

3Pos095 Mg ポリマー再考２

Revisiting "Mg-Polymer" 2

Mahito Kikumoto, Shuichi Takeda, Yuiticho Maeda (*Struct. Biol. Center, Grad. Sch. Sci., Nagoya-U.*)

In order to study the dynamic property of polymerized actin to clarify the relationship between polymorphism and actin state/structure, we have been searching for another state of the polymerized actin. When Physarum actin was polymerized in the presence of Mg^{2+}, the polymerized actin showed a low viscosity and a high ATPase rate. This state was designated "Mg-polymer" in 1967. Independently, it was found that rabbit muscle actin polymerized in the presence of β-actinin (= CP/CapZ) and Mg^{2+} has similar properties. We set out revisiting "Mg-polymer" to reidentify mechanistic, structural and biochemical bases of this state. Because of its high ATPase activity, this state may be a good model for analyzing the ATPase mechanism and/or polymorphism of the actin filament.

3Pos096 アクチンの集団運動内でのアクチン間の長距離相互作用

Long range interaction among of actin filaments under their collective movement

Yuto Fujita, Shigeru Sakurazawa (*Grad. Sch. System Info., Future Univ. Hakodate*)

Collective movements of actin filaments are occurred in in vitro motility assay system with high concentration of actin solution. It is consider that this collective movement is caused as the result of changing direction which is caused by actin filaments colliding to each other. This theory is based on assumption that actin filaments move on surface of myosin heads. In our observations, it was found that actin filaments in higher region slid along with actin filaments sliding on surface of myosin heads. This phenomenon suggests that an actin filament can interact with a myosin by not only binding but also long range interaction.

3Pos097 Inter-subunit coordination around a ring-shaped ATPase

Liqiang Dai, Jin Yu (*Beijing Computational Science Research Center*)

Ring-shaped NTPases assemble multiple subunits into ring-like structures. Currently we focus on studying the inter-subunit coordination in F1-ATPase ring. To probe how the sequential coordination arises without the central subunit, we started simulating independent chemical site reactions and then gradually added a variety of neighbor-site couplings in the stochastic simulations. We notice that the sequential hydrolysis emerges under fairly straightforward coupling patterns. Based on these clues, we perform atomistic molecular dynamics and coarse-grained simulations to enable targeted subunit conformational changes, and watch the neighbor-site responses dynamically or mechanistically. Comparative studies of a range of ring-shaped NTPases would be conducted afterward.

3Pos098 Dynamic instability of microtubules in a ROS free environment

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Microtubule (MT) plays essential roles in cell organization, orientation and cytokinesis. Precise regulation of MT dynamics is critically important for cells to successfully carry out the cellular events. Recently some chemical species e.g., reactive oxygen species (ROS) has been suspected to modulate MT dynamics in cells. However, no detail has been understood yet due to the complicated cellular environment, and also for the unavailability of any in vitro investigation. Here, by performing simple in vitro investigations we have unveiled the effect of ROS on MT dynamics. By studying the dynamic instability of MTs in a ROS free environment and comparing with that in the presence of ROS, we have unveiled that MT showed enhanced dynamics in the ROS free environment.

3Pos099 DNA programmed active self-organization of microtubules on kinesin

Jakia Jannat Keya[1], Daisuke Inoue[2], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. of Chem. Sci. and Eng., Hokkaido Univ.*, [2]*Faculty of Sci., Hokkaido Univ.*)

Self-assembly of biomolecular motor protein microtubule (MT) converting chemical energy to mechanical work-have unique advantages for integrated micro and biodevices. However, controlling the self-association storing huge amount of energy and localized dissociation of MT assembly concurrently is a challenging task. DNA, a programmable tool can control MT assembly due to its specific base pair interaction. Therefore in this work, we have demonstrated DNA programmed active self-organization (AcSO) of MTs on kinesin coated surface in vitro. MTs were modified by designed DNA sequences by click reaction. Self-organization as well as dissociation was performed by specific DNA sequences which will enable us to explore new space in the development of nanotechnology.

3Pos100 Observation of the gliding machinery of Mycoplasma mobile by Quick-Freeze Deep-Etch Replica Electron Microscopy

Clothilde Bertin, Yuhei O. Tahara, Tasuku Hamaguchi, Eisaku Katayama, Makoto Miyata (*Grad. School Sci., Osaka City Univ.*)

To clarify the surface protein assemblies of the unique gliding machinery of Mycoplasma mobile, quick-freeze deep-etch replication method was performed by using mica flakes as a solid support for washed cells. Leg-shaped structures sticking out from the cell surface and starfish-shaped structures were visualized. The leg-shaped structures would correspond to the leg protein. The starfish-shaped structures have been identified as the crank protein arranged in a folded-trimer and complexed with, presumably, MMOB1650, since such complex has been isolated in single protein experiment. Finally, the observation of cell attached to the mica surface by its leg-like structures shows that gliding units might work in a non-synchronized manner.

3Pos101 V1-ATPase の A3B3 固定子における鞭毛タンパク質 FliJ の回転軸機能

Rotor function of flagella protein FliJ in A3B3 of V1-ATPase

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V1-ATPase consists of a stator A3B3 and a rotor DF. The FliJ of bacterial flagella and V1-D share a coiled coil structure albeit there is no apparent sequence homology between them. In this study, we inquire whether the FliJ is able to function as a rotor in A3B3 or not, by producing a series of chimera rotor consisting of both FliJ and V1-D. Albeit the chimera rotor consisting of entire FliJ rarely formed the reconstituted complex with A3B3, we were able to find rotation molecules including the chimera rotor with a torque 1/2~2/3 of the WT. Our results indicate that the FliJ functions as the rotor in A3B3, that is, non of specific interaction between a rotor and stator hexamer is needed for producing the torque.

3Pos102 試験管内再構築系を用いた細胞膜-アクチンコーテックス複合体変形機構の解明

Reconstituted actomyosin cortex deformed in size dependent manner

Yukinori Nishigami[1], Hiroaki Ito[1], Seiji Sonobe[2], Masatoshi Ichikawa[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Sci., Univ. Hyogo*)

A cell shape plays an important role in variety types of biological phenomena. Actomyosin connects with the inner membrane and translates its contractile force to cause deformation of the cell. Because of the complexity of the force transduction between the cortex and the membrane, consistent mechanism explaining the macroscopic shape remains unclear. Here, we have developed a novel reconstituted system and surveyed the relation between curvature of the cortical membrane and its deformation mode. As a result, the characteristic deformation depended on the system size or curvature-radius. This curvature dependence was explained by the theoretical description including cortex elasticity and contractility.

3Pos103 高速原子間力顕微鏡を用いた回転軸の無い V_1-ATPase のコンフォメーション変化の観察

Observation of Conformational changes of rotorless V_1-ATPase using High-Speed Atomic Force Microscopy

Motonori Imamura[1], Kazuya Nakamoto[2], Takayuki Uchihashi[1,3,4], Takeshi Murata[2,5], Toshio Ando[1,3,4] ([1]*Dept. of Phys., Kanazawa Univ.*, [2]*Grad. Sch. Sci., Chiba Univ.*, [3]*Bio-AFM FRC, Kanazawa Univ.*, [4]*CREST, JST*, [5]*PRESTO, JST*)

V-ATPase regulates the pH level of intracellular compartments in our body by pumping H^+ through membranes driven by ATP. The structure and rotary mechanism of the V-ATPase is similar to F-ATPase. The hydrophilic domain of V-ATPase (V_1-ATPase) has the catalytic activity of ATP hydrolysis. The X-ray crystal structure of V_1-ATPase, derives from *Enterococcus hirae* (*E. hirae*), which pumps Na^+ instead of H^+, shows several conformations depending on the ATP/ADP binding. This study uses high-speed atomic force microscopy (HS-AFM) to elucidate the rotary mechanism of the A_3B_3 part of V_1-ATPase derived from *E. hirae*. We will discuss the insights into the rotary mechanism between the known model from the crystal structure and dynamic observation from HS-AFM.

3Pos104 アクチンフィラメントに対する HMM とコフィリンの相互排他的結合の定量的解析

A quantitative analysis of exclusive binding of HMM and Cofilin to actin filament

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Heavy Meromyosin (HMM) and Cofilin bind cooperatively to actin filaments, and form clusters on the filaments. Although we believe that the cooperative binding is achieved through conformational changes of actin subunits, which is evoked by binding of them, the physiological role of the cooperative interactions remains unknown. In this study, therefore, we examined what will happen if their two proteins were allowed to bind to actin filaments simultaneously as in live cells. To analyze the cooperative binding, HMM-GFP and Cofilin-mCherry were introduced into flow chambers that immobilized actin filaments, and observed by fluorescence microscopy. The quantitative analysis of fluorescence micrographs suggested that the two cooperative biding were mutually exclusive.

3Pos105 フォトクロミック分子で変化する有糸分裂キネシン Eg5 の ATP 分解活性経路の光制御ステップのストップドフロウ解析

Stopped flow analysis on the photo-regulated step in ATPase kinetic pathway of mitotic kinesin Eg5 modified with photochromic molecule

Kentaro Saito, Kei Sadakane, Yuki Tamura, Ryoma Yamamoto, Shinsaku Maruta (*Grad. Sch. Eng., SOKA Univ.*)

Previously we have demonstrated that the ATPase activities and microtubules gliding ability of the mitotic kinesin Eg5 mutants modified with photochromic molecules were regulated reversibly upon ultraviolet and visible light irradiations. In this study, we analyzed which step in ATPase kinetic pathway is regulated by photochromic molecules photo-reversibly with stopped flow apparatus utilizing fluorescent ATP analogues, Mant-ATP and NBD-ATP. The Eg5 mutants which have a reactive single cysteine residue were modified with thiol group reactive azobenzene or spiropyrane derivatives. The efficient FRET between 127W of Eg5 and Mant-ATP during hydrolysis of Mant-ATP was observed. Therefore, we utilized the FRET for the kinetic studies using Mant-ATP.

3Pos106 キネシン Eg5 の新規フォトクロミック阻害剤存在下における運動アッセイと速度論的解析

Kinetic analysis on the mitotic kinesin Eg5 ATPase and in vitro motility assay in the presence of a novel photochromic inhibitor

Kei Sadakane, Yuhki Tamura, Kentaro Saitoh, Ryoma Yamamoto, Shinsaku Maruta (*Soka University, Graduate school of Engineering, Division of Bioinformatics*)

Previously we have synthesized a novel photochromic potent inhibitor for mitotic kinesin Eg5, MASP-Cys (Maleimide spiropyran L-cysteine). Yamamoto et al. showed that the inhibitory effect of the MASP-Cys on the microtubule dependent ATPase activity of Eg5 were altered by the photo isomerization upon ultraviolet and visible light irradiations. In this study, we analyzed the effect of MASP-Cys on the ATPase kinetic pathway with stopped flow apparatus using fluorescently labeled ATP analogue, Mant-ATP. FRET between Mant group and the intrinsic Trp127 residue was observed. The rates of ATP biding and ADP release in the presence of the inhibitor were measured using the FRET. In vitro microtubule gliding assay of Eg5 was also studied in the presence of MASP-Cys.

3Pos107 好熱菌由来の回転モーター F_1 のカップリングスキーム

Coupling scheme of the rotary motor thermophilic F_1

Kengo Adachi[1], Kazuhiro Oiwa[2], Masasuke Yoshida[3], Kazuhiko Kinosita, Jr.[1] ([1]*Dept. Physics, Waseda Univ.*, [2]*Adv. ICT Res. Inst., NICT*, [3]*Dep. Mol. Biosci., Kyoto Sangyo Univ.*)

Thermophilic F_1 is a rotary molecular motor driven by sequential hydrolysis of ATP in three catalytic sites. In the standard coupling scheme, ATP binding starts rotation at 0°, and at ~200° the ATP is cleaved, and then the ADP is released around 240° after a third ATP is bound. The Pi release is at 200° or 320°, yet unsettled. Rates of ATP cleavage and Pi release in catalytic dwell were 2030 s^{-1} and 840 s^{-1}, whereas the two rates for ATPγS were 31 s^{-1} and 450 s^{-1}, respectively. ATPγS is thus a slowly hydrolyzed ATP analog and moreover thio-Pi release is about twice as slow as Pi release. With ATPγS mixed in ATP, the slow thio-Pi release could not be discerned at 320° after long dwells at 200° corresponding to ATPγS cleavage.

3Pos108 The structure of the flagellar filament of magnetotactic bacterium MO-1 by electron cryomicroscopy

Juanfang Ruan[1], Takayuki Kato[1], Keiichi Namba[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*QBiC, RIKEN*)

Many bacteria move by rotating a rigid, helical organelle, the flagellum. The bacterial flagellum is a motility apparatus and represents one of the most sophisticated nanomachines in biosphere. Recently we reported that the flagellum of MO-1, a marine magnetotactic bacterium, is a complex organelle consisting of 7 filaments and 24 fibrils that form a tight bundle enveloped by a sheath. The diameter of the gently curved filament is 12 nm, which is nearly half of that of Salmonella (23nm). Here we report the filament structure revealed by electron cryomicroscopy. The surface of the filament is smooth with much less helical feature than that of the Salmonella filament, but the subunit folds of the filament core and their helical arrangements appear to be very similar.

3Pos109 鞭毛内腕ダイニンの形成にはアクチンの N 末端側が重要である

N-terminal sequence of actin is critical for the assembly of flagellar inner-arm dyneins

Takako Kato-Minoura, Yuko Horikoshi, Kaori Imaeda (*Dept. Biol. Sci., Chuo Univ.*)

Flagellar inner-arm dyneins contain actin as a light chain. A *Chlamydomonas* mutant, *ida5*, lacks the conventional actin but expresses a divergent type of actin (NAP), and because of this lacks four of the seven kinds of inner arm dyneins. To elucidate the actin sequence crucial for dynein assembly, we examined the dynein-forming ability of chimeras of actin and NAP. Expression of a chimera comprised of an N-terminal actin portion and a C-terminal NAP portion in *ida5* recovered the missing species of inner arm dyneins and wild-type motility. In contrast, expression of a chimera of an N-terminal NAP portion and a C-terminal actin portion did not recover missing dyneins or motility. These results suggest the importance of the N-terminal sequence of actin for dynein assembly.

3Pos110 Single-molecule analysis of hybrid *Enterococcus hirae* V_1-ATPase toward elucidation of the chemo-mechanical coupling scheme

Yoshihiro Minagawa[1], Hiroshi Ueno[1], Hiroyuki Noji[1], Takeshi Murata[2], Ryota Iino[3,4] ([1]*Dept. App. Chem., Grad. Sch. Eng., The Univ. Tokyo*, [2]*Grad. Sch., Univ. Chiba*, [3]*Okazaki Inst. Integ. Biosci., NINS*, [4]*Grad. Univ. for Adv. Studies,*)

E. hirae V_1-ATPase(EhV_1) is a rotary molecular motor. To determine the absolute angle of each elementary reaction step of ATP hydrolysis occurring in a single catalytic subunit (A subunit) of EhV_1, we applied a hybrid EhV_1 that carries a single mutant A(F425E) subunit and 2 wild-type A subunits. The ATP binding and another elementary reactionrates of the A(F425E) were distinctly lower than those of the wild-type. Hybrid EhV_1 exhibited three pausing positions separated by 120º that had different duration times in the rotation. We identified absolute angles of two lengthened pauses corresponding to slow elementary reacton steps of A(F425E). As a result, after ATP binding at 0º, either one of ATP cleavage, ADP or Pi release occur at 240º in a single A subunit of EhV_1.

3Pos111 マイコプラズマ滑走タンパク質 Gli349 を構成するリピート断片の立体構造解析

Structural analysis of repeat fragments consisting of the gliding protein Gli349 from *Mycoplasma mobile*

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The leg protein Gli349 from *Mycoplasma mobile* is responsible for the gliding of the cell on a solid surface. Gli349 consists of 18 repeats of ~100 residues, but its detailed structure remains unknown. We previously found that the Gli349 fragment named KLM, which overlaps with the regions of repeats K, L, and M, is soluble, and small-angle X-ray scattering (SAXS) measurement revealed that KLM is composed of three globular domains, consistent with the tandem repeat structure of Gli349. Here, we searched for the repeat boundaries in the KLM fragment by trypsin limited proteolysis and TOF-MS analysis. Further fragmentation of KLM resulted in a single globular structure with a small tail, as revealed by SAXS, suggesting the detailed repeat boundaries in Gli349.

3Pos112 腸内連鎖球菌由来 V-ATPase の Na^+濃度依存の回転

Na^+-dependent rotation of *Enterococcus hirae* V-ATPase

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Recently, we established a single-molecule rotation assay for Na^+-transporting *E. hirae* V-ATPase (EhV_oV_1). Interestingly, with a load-free probe and at high Na^+ concentration, EhV_oV_1 rotated slower than EhV_1 with no clear three pauses observed for EhV_1, which suggests that EhV_o limits the rotation. However, no clear multiple pauses reflecting the 10-fold symmetry of V_o-rotor were resolved. So, to resolve the clear small pauses in EhV_oV_1, we observed the rotation of EhV_oV_1 at lower Na^+ concentration. At zero Na^+ concentration (contaminating ~10 μM Na^+), rotation rate of EhV_oV_1 decreased to the 17% of that at high Na^+ concentration (125 mM), suggesting that the Na^+-binding becomes rate-limiting. We are currently trying to determine the cause of this slow-rotation.

3Pos113 カーボンナノチューブを用いたべん毛モーターの抑制的制御

Carbon nanotube-based deactivation of bacterial flagellar motors

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We showed that carbon nanotubes (CNTs) work as a new platform to regulate activity of linear motors (Inoue et al., ACS Nano, 2015). Here our method was tested for bacterial flagellar motors. Multiwall CNTs were sparsely coated on the glass surface and the tethered rotation of E. coli cell was observed. Laser irradiation (1064 nm, 50 mW) to the CNTs induced a sudden stop of the rotation of the cells near the CNTs, whereas the direct irradiation to the cell did not change the rotation. In the presence of the oxygen-scavenging system, however, the deactivation was turned into the activation of the rotational speed from ~5 Hz to ~10 Hz, indicating that CNT-based deactivation as well as activation can be applied for various biomolecules including rotary motors.

3Pos114 ゆらぎ計測によるゼブラフィッシュ色素顆粒輸送の力-速度関係

Force-velocity relation of organelle transport in Zebrafish melanophores: new fluctuation analysis

Shin Hasegawa[1], Kazuho Ikeda[2], Yasushi Okada[2], Kumiko Hayashi[1] ([1]*Sch. Eng., Tohoku Univ.*, [2]*QBiC, RIKEN*)

Melanophores have melanosomes filled with melanin pigments. Melanosomes disperse in the cytoplasm or aggregate in the perinuclear region in response to hormones, that allowing *Zebrafish* to display color change. Melanosome transport by microtubule motors causes the dispersion and aggregation. In the previous study [K. Hayashi and Y. Okada, 2P145 (2013)], organelles in neulons were found to be transported by multiple motors. In our study, the cooperative transport of melanosomes in the melanophores was investigated. We observed melanosome's motion by brightfield transmission light microscopy. Analyzing the motion based on the fluctuation theorem of non- equilibrium statical mechanics, we discuss the force-velocity relation of the melanosome transport.

3Pos115 ダイニンによって駆動される微小管の polar あるいは nematic 集団運動
Polar or nematic motion of collective microtubules driven by dyneins

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Using in vitro motility assays, we have studied collective motion and emergence of patterns of microtubules (MTs) driven by dyneins. We found that MTs can display nematic order, millimeter-scale meandering streams or millimeter-scale vortices. To explore the conditions causing such phase-shifts, we use different types of dynein (dynein c, g of Chlamydomonas flagella and full-length cytoplasmic dynein expressed in HEK293 cells) and MTs with various mean lengths. In the motility assays, MTs gradually aligned and finally forms streams meandering across a very large distance or vortices. Variation in the time-course and MT-length dependence of pattern formation suggest that the vortex formation may reflect the mechanical properties of dyneins.

3Pos116 光応答性 DNA を用いた微小管集団運動の時空間制御
Spatiotemporally controlled collective motion of self-propelled microtubules by using photoresponsive DNA

Ryuhei Suzuki[1], Kyohei Uenishi[1], Daisuke Inoue[2], Kazuki Sada[1,2], Akinori Kuzuya[3], Hiroyuki Asanuma[4], Akira Kakugo[1,2] ([1]*Grad. Sch. Chem. Sci. Eng., Hokkaido Univ.*, [2]*Fac. Sci., Hokkaido Univ.*, [3]*Fac. Chem. Mater. Bioeng., Kansai Univ.*, [4]*Grad. Sch. Eng., Nagoya Univ.*)

Biomolecular motor system microtubule/kinesin has been considered a candidate for developing microdevices which can perform mechanical work by consuming chemical energy of adenosine triphosphate (ATP). Recently, we established a method to organize microtubules into assembled structure by utilizing DNA interaction, which was termed as active self-organization. In this work, we report spatiotemporal control of microtubules assembly formation using photoresponsive DNA which also allows controlling collective behavior of microtubules.

3Pos117 Formation and rupture of a motorized cytoskeletal network

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We established a set of microtubule-kinesin systems which exhibit various spatiotemporal patterns including the static network, active network, aggregation phase, and population of asters. They emerged according to the concentrations of the system components and the motor activities. A coarse-grained model was constructed and revealed two determinants for the formation of these spatiotemporal patterns: First, motor accumulation onto a number of interconnected high-density spots. Second, nonlinear dependence of the force between the spots on motor concentration. We also found that elastic energy stored in the networks can be transformed into mechanical work during network rupture. This mechanical feature might contribute to symmetry breaking in cell migration.

3Pos118 SH1 ヘリックス変異ミオシン II の運動活性と熱安定性
Motile activity and thermal stability of SH1 helix mutant myosin II

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Mutations at the SH1 helix of the myosin II motor domain, such as E706K (Glu-706 to Lys) and R702C (Arg-702 to Cys) have been reported to link to some autosomal-dominant diseases. The SH1 helix acts as a linker for transmitting the structural changes of ATP-binding site in the catalysis domain to the lever arm. To investigate the effects on the motile activity and the thermal stability, we introduced each of two mutations (E683K and R686C) into the SH1 helix in Dictyostelium myosin II. The mutations resulted in a decrease in the sliding velocity and the thermal stability, and the thermal aggregation. Therefore, it is suggested that the SH1 helix participates in the process of the sliding motion and is one of factors that determines the thermal stability of myosin II.

3Pos119 高速 AFM によるミオシン VI のモーターメカニズムの解明
Motor mechanism of myosin VI studied by high-speed AFM

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Myosin VI is the unique class of myosin that moves towards the minus end of actin filaments and known to move with a step size (~36 nm) larger than that expected from its canonical lever-arm length. Last year, we presented high-speed AFM movies that directly captured artificially dimerized myosin VI molecules moving wiggly towards the minus end of actin filaments with large and small strides. Here, to gain an insight into the motor mechanism of myosin VI, we analyzed the captured movies in detail, mainly about how myosin VI makes a large stride on actin filaments.In this presentation, we will report more details of the results.

3Pos120 Detection of TRPC and Orai1 proteins in bovine ciliary muscle cells prepared by a Percoll density-gradient centrifugation method

Motoi Miyazu, Kosuke Takeya, Toshiyuki Kaneko, Akira Takai (*Asahikawa Medical Univ.*)

In bovine ciliary muscle (BCM), stimulation of M_3-muscarinic receptors (M_3R) opens two types of non-selective cation channel with different unitary conductances which serve as major pathways for Ca^{2+} entry during sustained contraction. The molecular entities of these channels are still unknown. Recently we developed a new method to obtain BCM cells with unprecedented quality and amount and applied it to examine the existence and localization of TRPCs and Orai1. The ciliary body dissected from bovine eye were treated with collagenase, and the dispersed cells were subjected centrifugation through discontinuous Percoll density-gradient. Immunological staining revealed abundant expression of TRPC1, TRPC3, TRPC4, TRPC6 and Orai1 (as well as of M_3R) in the plasma membranes.

3Pos121 ストレス線維における張力依存的な ERK の活性化
Tension-dependent ERK activation on actin stress fibers

Hiroaki Hirata[1,2], Mukund Gupta[2], Sri Ram Krishna Vedula[2], Chwee Teck Lim[2,3], Benoit Ladoux[2,4], Masahiro Sokabe[2,5] ([1]*R-Pharm Japan*, [2]*Mechanobiol. Inst., Natl. Univ. Singapore*, [3]*Dep. Biomed. Eng., Natl. Univ. Singapore*, [4]*Univ. Paris Diderot*, [5]*Grad. Sch. Med., Nagoya Univ.*)

Activation (phosphorylation) of the MAP kinase ERK, which is involved in the regulation of diverse cellular functions, is largely affected by intracellular and extracellular mechanical cues. Here, we show that ERK is activated on actin stress fibers dependently upon tension developed in the fibers. While ERK was localized to the actin cytoskeleton, inhibition of myosin II decreased the amount of phosphorylated ERK, but not total ERK, on stress fibers. In myosin II-inhibited cells, stretching of extracellular substrates restored ERK phosphorylation on stress fibers. By quantifying myosin II- or stretch-mediated tensile force in individual stress fibers, we showed that larger tensile force in stress fibers caused more phosphorylation of ERK on the fibers.

3Pos122 表層ストレス応答を制御する膜内切断プロテアーゼ RseP による基質切断反応の生細胞内イメージング
Live-cell imaging of a proteolytic event by an intramembrane protease RseP that regulates extracytoplasmic stress response

Yohei Hizukuri, Yoshinori Akiyama (*Inst. Virus Res., Kyoto Univ.*)

The extracytoplasmic stress responses (ESR) play key roles to cope with cell surface stresses and are important for bacterial survival under stressed conditions and expression of virulence determinants. In the σ^E-pathway ESR, an extracellular stress signal is transduced into the cell interior across the cell membrane through sequential cleavages of anti-σ protein RseA by two membrane proteases DegS and RseP. We are trying to understand this dynamic cellular process by fluorescent microscopic observation and single cell imaging analysis. We observed RseP-dependent release of the RseA cytoplasmic domain from the membrane by using the GFP-RseA fusion protein in living cells. We are also visualizing and analyzing dynamic behavior of candidates of novel RseP substrates.

3Pos123 ハイドロゲルの表面力学場及び表面生化学の両条件に対する分散培養 iPS 細胞の増殖応答性
Proliferation response of dissociated iPS cells to the dual parameters of surface mechanics and biochemistry of culture hydrogel

Kenta Mizumoto[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Univ. Kyushu*, [2]*IMCE., Univ. Kyushu*)

Feeder-free dissociating culture of iPS cells has been required for stable and easier supply of them for further clinical applications. One of the problems with the dissociating culture is that iPS cells undergo apoptosis due to the loss of cell-cell adhesion and also affected by the strength of cell-substrate adhesion. To get insights into the optimized conditions for the dissociating culture of iPS cells, we systematically investigated the proliferation response of them to both conditions of surface mechanics and biochemistry using the laminin-fixed elasticity-tunable hydrogel. We discuss the contribution of the dual-parameters-regulated motility of iPS cells to assemble each other and to stabilize the colony, which is essential for high efficient proliferation.

3Pos124 Rotational diffusion of proteins in crowded environment: NMR spectroscopy and molecular dynamics simulation

Po-hung Wang[1], Isseki Yu[1], Hideyasu Okamura[2], Takanori Kigawa[2], Yuji Sugita[1,3,4,5] ([1]*RIKEN Theoretical Molecular Science Laboratory*, [2]*RIKEN Yokohama*, [3]*RIKEN AICS*, [4]*RIKEN QBiC*, [5]*RIKEN iTHES*)

The macromolecular volume fraction inside a cell can be up to 40% and has strongly affected biological processes, e.g. diffusion. Rotational diffusion in crowded media is decreased as viscosity increases. Our collaborators measured the relaxation times of chicken villin headpiece in concentrated solutions using NMR. Two rotational time scales were discovered, one of which was found to increase as the concentration increased. We performed simulations of the same protein in similar concentrations and computed the rotational diffusion coefficient. Our results showed a moderate agreement between calculation and experiment. The TIP3P water model tends to accelerate protein diffusion in dilute condition and to enhance protein-protein interaction in crowded environment.

3Pos125 フィラミンのアクチン結合ドメインは、極性をもった粘菌細胞後部のアクチンフィラメントを認識し特異的に結合する
Actin binding domain of filamin recognizes and specifically binds to posterior actin filaments in polarized *Dictyostelium* cells

Keitaro Shibata, **Taro Uyeda** (*Biomed. Res. Inst., AIST*)

The actin binding domain (ABD) of filamin was known to colocalize specifically with actin filaments in the rear of a polarized *Dictyostelium* cell. However, how the ABD interacts with a specific subset of actin filaments was unknown. Here, we followed translocation of filamin ABD fused with photoswitchable fluorescent protein *in vivo*. We found that the majority of filamin ABD in the middle of an elongated cell rapidly diffused in the cytosol and specifically bound to posterior actin filaments. In contrast, actin filaments abundant in the anterior lamellipodia were hardly bound with filamin ABD. We suggest that the posterior actin filaments are structurally different from the anterior filaments and filamin ABD recognizes the structure of the posterior filaments.

3Pos126 心室・心房の組織片による自律拍動の同期化
Synchronization of spontaneous beating of tissue fragments of atrium and ventricle

Ryuichi Shinozaki, Tomonori Takahashi, Yuichi Asanuma, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

The heart consists of upper chambers, atrium and lower chambers, ventricle and their mechanical-electrical properties differ in order to pump blood. Microscopically, the mechanical-electrical properties of cardiac myocytes, muscle cells, from these chambers also differ. However, macroscopically, the properties of tissue fragments from different chambers at sub millimeter scale have not well investigated including their beat activity. We have focused on studying the spontaneous contraction activity of sub mm tissue fragments and their coupled oscillations and synchronization by attaching multi fragments. We present the contraction activity, such as interbeat intervals of fragments and the synchronization probabilities between fragment pairs from different regions.

3Pos127 細胞サイズ球状閉鎖空間内でのアクトミオシンリングの自発形成と収縮

若手招待講演

Cell-sized spherical confinement induces the spontaneous formation of contractile actomyosin rings *in vitro*

Makito Miyazaki[1], Masataka Chiba[1], Hiroki Eguchi[1], Takashi Ohki[1], Shin'ichi Ishiwata[1,2] ([1]*Dept. of Physics, Waseda Univ.*, [2]*WABIOS, Waseda Univ.*)

During cell division, many animal cells transform into a spherical shape and assemble a cytokinetic ring to separate the cell body into two. Here, we demonstrated *in vitro* that actin polymerization inside cell-sized spherical droplets induced the spontaneous formation of single ring-shaped actin bundles in the presence of bundling factors. Despite a lack of spatial regulatory signals, the rings always assembled at the equator to minimize the elastic energy of the bundles. Myosin promoted ring formation by the dynamic remodeling of actin networks, and an increase in the effective concentration of myosin triggered ring contraction. These results will help us understand how animal cells coordinate cell shape and actomyosin activities to direct cytokinesis.

3Pos128 大腸菌走化性シグナル伝達タンパク質の極局在と細胞内シグナル伝達の関係

Relationship between polar localization of chemotactic proteins and intracellular signaling under steady-state of *Escherichia coli*

Yong-Suk Che[1], Hajime Fukuoka[1], Yuichi Inoue[2], Hiroto Takahashi[2], Akihiko Ishijima[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ*, [2]*IMRAM, Tohoku Univ*)

In chemotaxis system, phosphorylated CheY (CheY-P) works as intracellular signaling molecule, and CheY-P is generated and erased by polar localized CheA and CheZ, respectively. We recently proposed, under steady-state, the CheY-P concentration is dynamically changed to coordinate the switching between flagellar motors. In this study, to understand the role of polar localization of CheA, CheZ, and CheY for the signaling under steady-state, we measured switching coordination between motors in the absence of CheA-, CheZ-, CheY-localization. In the absence of CheZ-localization, two motors coordinate switched as wild-type cell. However, in the absence of CheA-, and CheY-localization, the switching coordination was absent. We will discuss about these results in the meeting.

3Pos129 バクテリア FlgN シャペロンの機能構造スイッチの分子基盤

Molecular basis for a structural switch of FlgN that regulates its chaperone activity

Miki Kinoshita[1,2], Yuki Nakanishi[2], Yukio Furukawa[1], Katsumi Imada[2], Keiichi Namba[1,3], Tohru Minamino[1] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ*, [3]*QBiC, RIKEN*)

A bacterial flagellar chaperone FlgN binds to the hook-filament junction proteins FlgK and FlgL in the cytoplasm and escorts them to the sorting platform made of a membrane protein FlhA. FlgN also binds to a cytoplasmic export protein FliJ during the export process. FlgN consists of three α-helices, α1, α2 and α3. α3 helix of FlgN is responsible for the interactions with FlgK/L and FliJ. But, it remains unknown how FlgN induces the association and dissociation of its binding partners. In this study, we analyzed the effect of deletion of a loop between α1 and α2 of FlgN on the interactions with FlgK and FliJ. We will show that this loop controls the binding affinities of FlgN for FlgK and FliJ and discuss the regulatory mechanism of the FlgN chaperone activity.

3Pos130 アクチン結合タンパク質によって変化するアクチン繊維内モノマーの位置ゆらぎ

ABPs alter the fluctuations of monomer configurations within an actin filament

Hirotaka Ito[1], Kohei Monma[1], Sakura Maesato[1], Kenji Kobayashi[1], Ryoki Ishikawa[2], Hazime Honda[1] ([1]*Dept. of Bioneg., Nagaoka Univ. Tech*, [2]*Gunma Pref. Col. Health Sci.*)

Supramolecular structures of an actin filament may be considered to be not so strictly defined in solution as suggested by electron micrographic or crystallographic studies. In order to quantify the structural fluctuations of actin filament upon binding various actin-binding proteins (ABPs), we have tried to measure the fluctuations of the signal from inter-protomer FRET (Fluorescence Resonance Energy Transfer) within single actin filaments under fluorescent microscope. The signals were distributed into 4-classes, and were shifted upon binding of myosin, tropomyosin or drebrin in a dose dependent manner. The fluctuations may play an important role in the binding of ABPs.

3Pos131 変異リアノジン受容体における分子動力学解析とカルシウムシグナル可視化解析の相関

Correlation of molecular dynamics analysis and calcium signaling in mutant ryanodine receptors

Toshiko Yamazawa[1], Takashi Murayama[2], Hideto Oyamada[3], Junji Suzuki[4], Nagomi Kurebayashi[2], Kazunori Kanemaru[4], Maki Yamaguchi[1], Shigeru Takemori[1], Katsuji Oguchi[3], Takashi Sakurai[2], Masamitsu Iino[4] ([1]*Dept Mol. Physiol., Jikei Univ. Sch. Med.*, [2]*Dept. Pharmacol., Juntendo Univ. Sch. Med.*, [3]*Dept. Pharmacol., Sch. Med., Showa Univ.*, [4]*Dept. Pharmacol., Grad. Sch. Med., The Univ. Tokyo*)

Ryanodine receptors (RyRs), located in the endoplasmic reticulum (ER) membrane, are required for intracellular Ca^{2+} release. Mutations in RyR1 can lead to severe genetic conditions that affect skeletal muscle, e.g., malignant hyperthermia (MH). We investigated properties of the RyR1 channels carrying disease-associated mutations at the N-terminal region. HEK293 cells expressing the mutant RyR1 channels exhibited alterations in Ca^{2+} homeostasis, i.e., enhanced caffeine sensitivity, decrease of ER Ca^{2+} contents, increases in resting cytoplasmic Ca^{2+} concentration, changes in pattern of electrostatic interaction. These results suggest that exploration of the functional mutations of RyR1 is probably effective in preventive diagnosis of patients associated with MH disease.

3Pos132 再構成系を用いた WAVE 複合体制御分子機構の解明

Reconstitution of the WAVE complex regulation mechanism

Tomotaka Komori[1,2], Scott Hansen[2], R. Dyche Mullins[2] ([1]*Univ. of Tokyo*, [2]*UCSF*)

The WAVE regulatory complex (WRC) is the main nucleator of the Arp2/3 complex, a key regulator for actin filament branching in lamellipodia. WRC is maintained in an autoinhibited state by preventing its VCA domain from being exposed outside of the complex to prevent Arp2/3 complex activation. The autoinhibition is thought to be regulated by small GTPase binding and phosphoregulation by kinases. In contrast, little is known about the mechanism activating WRC. To tackle this issue, we reconstituted WRC using purified proteins and lipid coated beads. At the meeting, we will talk about this model and its application to study WRC activation.

3Pos133 超解像イメージングを用いた単離ミトコンドリアの膜構造観察

Super-resolution Imaging of Isolated Mitochondria with Structured Illumination Microscopy

Takahiro Shibata[1], Saki Yamashita[1], Kaoru Katoh[2], Yoshihiro Ohta[1] ([1]*Grad. Sch. Life Sci. & Bio Tech., TUAT*, [2]*AIST*)

Isolated mitochondria serve as a more easily controllable model system than intracellular mitochondria. Although mitochondria are typically isolated by cell homogenization, such treatment is potentially damaging to mitochondria. Therefore, we isolated mitochondria from cells without homogenization and observed the intactness of fluorescently-labeled outer and inner membranes of the mitochondria with SIM. The super-resolution imaging with SIM revealed that the outer and inner membranes mitochondria isolated without homogenization were intact and that the size of the mitochondria were similar to the intracellular mitochondria. On the other hand, mitochondria isolated with conventional procedures seemed to be fragmented during homogenization.

3Pos134 機能的な FRET プローブを用いた一細胞における大腸菌走化性受容体活性の検出

The detection of chemoreceptor cluster's activity in a single *E. coli* cell by the functional FRET probe

Tomoko Horigome[1], Hajime Fukuoka[2], Hiroto Takahashi[3], Yuichi Inoue[3], Akihiko Ishijima[2,3] ([1]*Grad. Sch. Life Sci., Tohoku Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*IMRAM, Tohoku Univ.*)

E.coli migrates to favorable environments by chemotaxis signaling system. To understand chemotaxis system as the behavior of protein in cytoplasm, the activity of chemotaxis proteins and the cellular response should be measured simultaneously. In this study, we are trying to develop functional FRET probe to measure the activity of chemoreceptor cluster as FRET between chemotactic proteins. Fluorescent Protein (FP)-fusion of CheW (CheW-FP) was functional, but FRET was not detected. FRET between CheY-FP and CheZ-FP appeared to be detected, while their function reduced. On the other hand, we succeeded in the construction of full-functional CheA-FP, therefore, we are now trying to measure FRET between CheA-FP. We would like to discuss about results at the annual meeting.

3Pos135 SOS を介した Ras positive feedback 制御から見た SOS Noonan 症候群変異体の分類

Classification of molecular dynamics in SOS Noonan syndrome mutants from the properties of SOS-mediated RAS positive feedback

Yuki Nakamura[1,2], Kayo Hibino[3], Yasushi Sako[1,2] ([1]*RIKEN*, [2]*Osaka university*, [3]*NIG*)

Noonan syndrome (NS) is a congenital and genetic disorder with various diseases. The 13% of NS patients has mutations in Son of sevenless (SOS). SOS is one of Ras guanine nucleotide exchange factor (GEF) and GEF activity of SOS is stimulated by interaction with the active form of RAS (positive feedback). Although NS mutations are located in various SOS domains, that NS mutants of SOS have abnormal GEF activity in common. It is unclear what causes abnormal GEF activity in each mutation. In this study, we observed TMR-conjugated SOS molecules in living HeLa cells using single-molecule imaging and analyzed the kinetics of SOS activation. Our observation indicated that mechanism caused SOS abnormal function and the role of positive feedback was different each SOS mutants.

3Pos136 単一心筋細胞の細胞外電位計測

Measurement of extracellular potential in single cardiomyocyte

Jyunpei Shimada[1], Kenji Yasuda[2], **Tomoyuki Kaneko**[1] ([1]*LaRC, Dept. Frontier Biosci., Hosei Univ.*, [2]*IBB, TMDU*)

Cardiomyocytes are known to change the membrane potential with spontaneous excitation. To measure the extracellular potential change in single cardiomyocytes, we developed the micro-electrode array (MEA) chip consisted of 10 μm diameter and 3 μm height of Pt-black-coated ITO electrode with agarose microchambers. Single cardiomyocytes were arranged one by one on this MEA chip with a micropipette. The extracellular potential in a single cardiomyocyte could be measured with this MEA chip. The amplitude of the extracellular potential change in a single cardiomyocyte was very small (<100 μV) and the beating rate was fluctuated. These results would promote to measure the membrane potential changes of single cardiomyocytes in cellular networks.

3Pos137 原子間力顕微鏡によるホヤ初期発生胚の弾性率の時空間測定

Spatial-temporal change in elastic modulus of ascidian embryo during an early stage of development by atomic force microscopy

Yuki Fujii[1], Wataru Koizumi[2], Kohji Hotta[2], Kotaro Oka[2], Takaharu Okajima[1] ([1]*Grad. Sch. Inform. Sci and Tech., Univ. Hokkaido*, [2]*Grad. Sch. Biosci. and Bioinfo., Univ. Keio*)

During developmental processes of embryo, the cell division and the configuration are highly organized and synchronized. Recent studies indicate that internal stress and strain of cells are essential for the formation of the self-organized cell structures [1]. However, little is known how the cell stiffness changes during the developmental process. In this study, we investigate the elastic modulus of the ascidian embryo during an early stage of development at the single cell level by atomic force microscopy (AFM). We found that the elastic modulus of not only dividing cells but also the surrounding cells synchronously changes according to the cell division.

[1] J. H. Shawky, et al. Developmental Biology 401 (2015) 152-164

3Pos138 接着分子 CADM1 による膵島 α 細胞グルカゴン分泌調節機構の解明

Cell adhesion molecule 1 (CADM1) regulate glucagon secretion in pancreatic α cells

Satoru Yokawa[1,2], Ryousuke Oguri[2], Yoshikazu Inoh[2], Ryo Suzuki[2], Tadahide Furuno[2], Naohide Hirashima[1] ([1]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [2]*Sch. Pharm., Aichi Gakuin Univ.*)

Pancreatic α cells in the islet of Langerhans secrete the glucagon, which elevates the glucose concentration in the blood. We previously showed that cell adhesion molecule 1 (CADM1) expressed in αTC6 cells, a murine α cell line, and mediated the cell-cell interaction. In this study, we found that CADM1-knockdown αTC6 cells decreased the glucagon secretion by arginine stimulation (44.8% compared to wild type cells), whereas average velocity of granule movements was similarly between them in Spinning-disk confocal microscopy. These results suggested that CADM1 regulated the exocytotic pathway such as the membrane fusion rather than the granule mobility.

3Pos139 HubP は FlhG を極に局在させることで海洋性ビブリオ菌のべん毛本数を制御する

HubP regulates the flagellar number by localizing FlhG at the cell pole in marine *Vibrio*

Norihiro Takekawa, Soojin Kwon, Seiji Kojima, Michio Homma (*Div. of Biol. Sci., Grad. Sch. of Sci., Nagoya Univ.*)

Marine bacterium *Vibrio alginolyticus* has a single polar flagellum, whose number is regulated positively by FlhF and negatively by FlhG. FlhF and FlhG localized at the pole of the cell, and the balance of the localization amount of these two proteins determines the flagellar number to be exactly one. Here we focus on HubP, a membrane protein that localizes at the cell pole and works as a "hub" for the positioning of various proteins at pole. We found that a deletion of *hubP* increased the flagellar number. By the deletion, the polar localization of FlhG-GFP was disrupted, whereas that of FlhF-GFP was not changed. Thus, HubP may regulate the balance of the FlhF and FlhG at the cell pole to determine the flagellar number.

3Pos140 ビブリオ菌 PomB 変異に対するべん毛モーター機能へのセリンの影響

Effect of serine on the flagellar motor function of a PomB mutant in *Vbrio alginolitycus*

Tatsuro Nishikino, Yasuhiro Onoue, Norihiro Takekawa, Shiwei Zhu, Seiji Kojima, Michio Homma (*Nagoya University Graduate School of Science*)

Many motile bacteria rotate flagella to swim toward a favorable condition. To generate torque, the stator complex, composed of PomA and PomB in *Vibrio alginolyticus*, interacts with the rotor. In our previous report, it is suggested that a conformational change in the periplasmic region of PomB is induced when the stator complexes are incorporated into the motor. Here, we found that the PomB-L160C/I186C mutant, which lost motility due to interference of the suggested conformational change with the formation of a disulfide bridge, restored motility in addition of Serine. Serine is known to be a chemoattractant and change the direction of flagella rotation. We speculated that serine affected the stator and the rotor interaction to suppress the defect.

3Pos141 細胞性粘菌における高感度膜電位イメージング

High-sensitivity fluorescence imaging of membrane potential in *Dictyostelium*

Yusuke V. Morimoto[1], Masahiro Ueda[1,2] ([1]*QBiC, RIKEN*, [2]*Grad. Sch. Sci., Osaka Univ.*)

The cellular slime mould *Dictyostelium discoideum* is a model organism for studies on cell motility, chemotaxis and differentiation. Chemotactic stimulation by cAMP was reported to elicit an influx of Ca^{2+} into the cell cytoplasm. This suggests that the membrane potential is changed by cAMP stimulation. However it remains unknown how the membrane potential works in cell motility and signal transduction. To investigate the role of membrane potential, we observed *Dictyostelium* cells stained with the membrane potential sensitive fluorescent dye by fluorescence microscopy. Then we observed periodical membrane potential changes depending on the spontaneous cAMP oscillations. We will discuss the role of membrane potential in the cellular function.

3Pos142 固体 NMR と MD シミュレーションによる抗菌ペプチド ボンビニン H2 および H4 の DMPC 膜結合構造の解析

Membrane binding structure of Bombinin H2 and H4 peptides in DMPC bilayers as studied by solid-state NMR and MD simulation

Izuru Kawamura[1], Yuki Kitahashi[1], Namsrai Javkhlantugs[1,2], Nyamsambuu Altannavch[2], Kazuyoshi Ueda[1], Akira Naito[1] ([1]*Grad. Sch. Eng., Yokohama Natl. Univ.*, [2]*Natl Univ. Mongolia*)

Bombinin H2 and H4 varying in the 2nd residue with L-Ile in H2 and D-allo-Ile in H4 are cationic antimicrobial peptides from frog skin of *Bombina variegata*. We have performed CD experiment, ^{13}C and ^{31}P solid-state NMR measurements and molecular dynamics (MD) simulations to investigate the structure of H2 and H4 in dimiristoylphosphatidylcholine (DMPC). CD spectra showed that both peptides formed mainly alpha-helical secondary structure in the membrane. ^{31}P NMR spectra showed that both H2 and H4 have similar strong interactions. In MD simulations, it is main difference that the interactions of N-terminal 1st and 2nd residues with lipid head groups at last 10 ns. Therefore, these results indicated that H4 has a specific interaction of N-terminus with lipid head groups.

3Pos143 オクタアルギニンの膜透過促進効果を示す曲率誘導性ペプチド

Curvature Inducing Peptides Accelerating Membrane Translocation of Octaarginine (R8)

Tomo Murayama, Shiroh Futaki (*ICR, Kyoto Univ.*)

Arginine-rich peptides, including R8, are known as representative cell-penetrating peptides. We previously reported that an N-terminal segment of epsin-1 possesses positive membrane curvature inducibility and that this peptide promotes membrane translocation of R8 (Pujals et al. ACS Chem. Biol., 8, 1894 (2013)). We this time assessed the curvature inducibility of amphipathic peptides, which are derived from proteins to induce tabulation/fusion of liposomal membranes. We also studied their effects on membrane translocation of R8, together with the lipid packing state under the treatment with these peptides.

3Pos144 ラマン分光によるスフィンゴミエリンの膜分布に関する研究

Sphingomyelin distribution in model membranes by Raman Spectroscopy

Koichiro Shirota[1], Kiyoshi Yagi[2], Takehiko Inaba[1], Pai-Chi Li[2], Yuji Sugita[2], Toshihide Kobayashi[1] ([1]*LBL, RIKEN*, [2]*TMSL, RIKEN*)

Sphingomyelin (SM) is a major sphingolipid in mammalian cells that forms specific lipid domains in combination with cholesterol. SM is miscible with dipalmitoylphosphatidylcholine (DPPC), whereas it is immiscible with dioleoylphosphatidylcholine (DOPC) and thus forms clusters. Experimental Raman spectra of SM, SM/DOPC and SM/DPPC liposomes show significant difference at approximately 1645 cm^{-1}, suggesting that the environment of the amide group of SM varies in these liposomes. We then identified this characteristic Raman band as amide I of the SM nanocluster with molecular dynamics simulation and density functional theory calculation. Consequently, we conclude that this amide I Raman band can be utilized to examine the membrane distribution of SM.

3Pos145 合成生物学研究のための一枚膜ベシクル内 DNA コンピュータ基盤遺伝子調節システムの開発

Development of a DNA computer-based gene-regulatory system confined in a giant unilamellar vesicle for synthetic biology research

Koh-ichiroh Shohda[1], Toru Nishikata[1], Yutetsu Kuruma[2], Akira Suyama[1] ([1]*The University of Tokyo*, [2]*Tokyo Institute of Technology*)

We developed a molecular system by which functions of an artificial genetic network was tested on in vitro level for synthetic biology research. The system consisted of a DNA computer-based gene-regulatory module, a cell-free protein synthesis system, and a cell-sized giant unilamellar vesicle (GUV). Furthermore, the DNA computer-based gene-regulatory module in GUV was directly controlled with a small molecule added from exterior of GUV. Compared with usual in vitro experiments, our system has a close environment to living cells. However our system never receive interferences from the genetic network of living cells. Therefore our system is suitable to examine intrinsic functions of artificial genetic networks designed in silico.

3Pos146 長鎖リン脂質と短鎖リン脂質で構成される脂質多成分系の相挙動と構造変化

The phase behavior and the structural changes of lipid multi-component system consisting of long- and short-chain phospholipids

Ryota Kobayashi, Tetsuhiko Ohba (*Dept. of Phys., Tohoku Univ.*)

Mixtures of long- and short-chain phospholipids show complicated phases depending on mixing ratios, total lipid concentration, and temperature. For example, DMPC/DHPC mixtures are transparent or turbid, and have high viscosity at intermediate temperatures. The structure at this range is not yet clear although some models such as wormlike micelles or elongated bicelles had been proposed.
In this study, when we introduce the packing parameter to describe this structure, it is turned out that there are few contradictions in case of applying to a lipid having a bulky head group or double chains.
We will discuss the structure and the driving forces that cause the structural changes together with the data of differential scanning calorimetry and fluorescence spectroscopy.

3Pos147 膜タンパク質の機能構造解析を指向した安定かつサイズ制御可能なナノディスクの開発

Development of stable and size-controllable nanodiscs for biophysical analysis of membrane proteins

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Nanodiscs (NDs) are lipid-polypeptide complexes applicable to solubilizing membrane proteins (MPs) in aqueous solution with maintaining native lipid bilayer environments. We developed a peptide forming NDs for analysis of MPs with various sizes and stabilities. We synthesized a 20-residue peptide, 20A, by modifying the ND-forming model peptide, 18A. NDs are made by mixing POPC with the peptides. We revealed that the sizes of 20A-NDs as well as 18A-NDs are controlled by lipid/peptide molar ratio. Moreover, 20A-NDs are much more stable than 18-NDs at high temperatures or high urea concentrations. The high stability of 20A-NDs is likely due to a polymerization of 20A by native chemical ligation.

3Pos148 アクチン重合によって引き起こされるリポソームの変形に与えるリン脂質組成の影響

Effect of lipid composition on the actin polymerization-driven shape change of giant liposomes

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Previous studies demonstrated that giant liposomes (GLs) changed to flat a disk- or paddle-like shape resulting from polymerization of the encapsulated actin. The effects of addition of actin-binding protein such as cross-linking factor on the deformation of actin-containing liposome have been studied. However, that of lipid composition remained unclear.
Actin-containing liposomes examined here were prepared by natural swelling from DOPC and DOPG, which have uniform hydrophobic tail. They changed to steric shapes, for an example, rugby ball-like shape, depending on actin concentration. We will observe localization of actin in GLs using fluorescence labeling, and will compare shape changes of GLs when other methods than natural swelling are adopted to prepare them.

3Pos149 巨大細胞膜小胞をマニピュレーションするための新規デバイスの開発

Development of new device for manipulation of giant plasma membrane vesicles

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Giant plasma membrane vesicles (GPMVs) isolated directly from cultured cells membrane is known for a useful model membrane. A protocol reported previously produces efficient yields of large (up to 10 μm) GPMVs. Now, we have developed a new method for manipulating of the GPMVs on micro holes of quartz plate to observe a specific GPMV for long time by optical microscopic techniques. We have prepared quartz plate with micro-holes (φ 2,3μm) and succeeded in manipulating the GPMVs on the micro-holes by a suction of the solution. The suction force was quite critical for the manipulation. The suction force was increased, the GPMVs sucked in the suction holes were pass through the micro-holes. We will present more details about suction conditions.

3Pos150 抗菌ペプチド・PGLa が誘起する脂質膜中のポア形成とマガイニン２との相乗効果

Antimicrobial Peptide PGLa-Induced Pore Formation in Lipid Membranes and its Synergistic Effect with Magainin 2

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To elucidate the pore formation induced by PGLa and its synergistic activity with magainin 2, we investigated interaction of PGLa with DOPG/DOPC-membranes using the single giant unilamellar vesicle (GUV) method. We observed that a gradual leakage of calcein from a GUV started stochastically and it completed without any changes in phase contrast image of the GUV. Based on its statistical analysis the rate constant (k_p) of the PGLa-induced pore formation was determined. These characteristics are the same as that of magainin 2 [1]. The k_p values of magainin 2-induced pore formation were elevated by the presence of low concentrations of PGLa which cannot induce pore formation. We discuss the mechanism of its synergistic activity.
[1] J. Phys. Chem. B 113, 4846, 2009.

3Pos151 Activation energy of the tension-induced pore formation in lipid membranes

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To understand the mechanism of tension-induced pore formation in biomembranes, it is important to obtain the values of its energy barriers. For this purpose, we investigated the temperature dependence of rate constant, k_p, of pore formation in lipid membranes of giant unilamellar vesicles under constant applied tension, σ, using the standard micropipette aspiration method. The values of k_p were determined by the method described in our previous paper [1]. Based on the analysis of the results we obtained the activation energy, U_a, of pore formation in membranes as a function of σ. The data was well fit by $U_a = U_0 + C/\sigma$, where U_0 and C are constants. We discuss this results based on the theory of the pore formation.

[1] Langmuir 29, 3848, 2013.

3Pos152 Effects of Line Tension on Antimicrobial Peptide Magainin 2-Induced Pore Formation

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To elucidate the effects of line tension of lipid membrane on magainin 2 (M2)-induced pore formation, we investigated the effect of cholesterol on the pore formation using single giant unilamellar vesicle (GUV) method. It is well known that the line tension increases as cholesterol conc. increased. The rate constant of M2-induced pore formation decreased with the increase in cholesterol conc. and at high conc. of cholesterol the pore formation was inhibited. On the other hand, in the constant tension-induced pore formation, high conc. of cholesterol decreased the fraction of ruptured GUVs. M2-induced area change of lipid membrane [1] was decreased in the presence of cholesterol. We analyze the effect of line tension based on the theory.

[1] Langmuir 31, 3391, 2015.

3Pos153 膜タンパク質-脂質相互作用の定量的解析法の開発と応用
Quantitative analysis of protein-lipid interactions

Takaharu Mori, Yuji Sugita (*RIKEN*)

Membrane proteins interact with surrounding lipid molecules when they act. To explore protein-lipid interactions at the atomic level, we proposed two methodologies: Voronoi-tesselation Monte Carlo integration method and Surface-tension replica-exchange molecular dynamics method. The former allows us to analyze the area per lipid around membrane proteins, and the latter is a new generalized-ensemble method for efficient conformational sampling of biological membrane systems. We applied our method to several membrane protein systems using our new MD software GENESIS. We observed changes in the area per lipid coupled with the conformational change of SecY and also succeeded to calculate free energy profile of membrane proteins.

3Pos154 脂質二重膜の流動性へのグラフェン酸化物の影響
Effect of graphene oxide substrate on the fluidity of lipid bilayer membrane

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Artificial lipid bilayers are valuable systems to study the fundamental interaction of chemicals and nanomaterials with plasma membrane. It is important to develop a new method using the function of nanomaterials. Recently, graphene oxide (GO) is applied to the biosensing because of its electric property or fluorescence quenching ability. We formed a supported lipid bilayer (SLB) on GO which was deposited on a SiO2/Si substrate with the vesicle fusion method, and evaluated the fluidity of SLB. We performed single particle tracking using quantum dots (Qd) to overcome fluorescence quenching by GO. We compared the diffusion of the same Qd-tagged lipid in the GO and SiO2 regions, and found that the lipid diffusion was slower in the GO region than that in the SiO2 region.

3Pos155 Conformational Control of Voltage Sensor Domains

Morten Bertz, Kazuhiko Kinosita, Jr. (*Waseda University, Dpt. of Physics*)

Voltage gating - the reaction of ion channels on response to changes in membrane potential - is fundamental to signal transduction in living organisms. In many ion channels, voltage sensor domains containing conserved positively charged residues move according to the transmembrane electrical field, and the resulting conformational change opens or closes the channel pore. The extent and direction of this movement, however, remain controversial, with both small translations and large-scale movements reported in the literature. Here, we attempt the manipulation of voltage sensor conformation using engineered probes as a substitute for voltage to shed light on the transitions involved in voltage gating.

3Pos156 電位依存性プロトンチャネルの 2 量体と単量体間のゲーティング特性の違いを増大させる変異体の解析
Mutation of a hydrophobic residue in S4 enhances the difference between monomeric and dimeric voltage-gated proton channels

Akira Kawanabe, Yasushi Okamura (*Osaka University*)

Voltage-gated proton channel (Hv1/VSOP) consists of four transmembrane helices (S1-S4) which play dual roles of voltage sensing and proton permeation. Hv1/VSOP assembles as a dimer to regulate the cooperative gating. Recent X-ray structure of Hv1/VSOP (Takeshita et al. 2014) suggested a hydrophobic layer in the channel. Mutant of L197C in the hydrophobic layer showed a large negative shift of the I-V relationship. Interestingly, the monomeric channel with the same mutation showed less shift of the I-V relationship, suggesting that environments that affect the gating are different in monomeric and dimeric channels.

3Pos157 イオン透過と選択性を記述する速度論モデル
A kinetic model describing punch-through of Na^+ through KcsA potassium channel

Takashi Sumikama, Kenichiro Mita, Shigetoshi Oiki (*Univ. of Fukui*)

Selective ion permeation through channels has been studied electrophysiologically through measurements of the reversal potential. In potassium channels, shifts of the reversal potential upon replacement of ion species are undetectable because of their strict selectivity to K^+ over other monovalent cations. Thus, an alternative method named the punch-through experiments has been applied for the KcsA potassium channel, in which single-channel currents in existence of Na^+ are evaluated in a wide voltage range. Here we developed a kinetic model, which successfully fits the punch-through currents. The model also predicts the single-channel current amplitudes when the ratio of intracellular Na^+ increases.

3Pos158 Proton transfer between cytochrome oxidases and the ATP synthase: examining the role of the membrane environment

Duncan McMillan[1,2], Sophie Marritt[3], Mengqui Li[2], Sune Jorgensen[4], Rikya Watanabe[1], Nikos Hatzakis[4], Julea Butt[3], Lars Jeuken[2], Gregory Cook[5], Hiroyuki Noji[1] ([1]*Department of Applied Chemistry, University of Tokyo, Tokyo, Japan*, [2]*School of Biomedical Sciences, University of Leeds, Leeds, UK*, [3]*School of Chemistry, Norwich Research Park, University of East Anglia, UK*, [4]*Nanoscience Center, University of Copenhagen, Copenhagen, Denmark*, [5]*Department of Microbiology and Immunology, University of Otago, Dunedin, New Zealand*)

Membrane-bound enzymes utilizing proton/electron transfer play a central role in the respiration of all life on earth. Quinone oxidoreductases generate transmembrane proton (ΔpH) and electrical gradients ($\Delta\Psi$), which in turn powers the ATP synthase, the key enzyme in producing chemical cellular energy (ATP). Little is known about the transfer of protons between oxidoreductases and the ATP synthase. The simultaneous analysis of proton and electron movement in respiratory chains is complex, requiring unique state-of-the-art biophysical technologies such as artificial bilayers, bioelectrochemistry and single-molecule microscopy. However, using a multidisciplinary approach this is now possible, and the studies underlining proofs of concept are presented.

3Pos159 反転膜ベシクルを用いたべん毛軸構造の構築
Construction of the flagellar axial structure using inverted membrane vesicles

Chinatsu Tatsumi[1], Hiroyuki Terashima[1], Tohru Minamino[2], Katsumi Imada[1] ([1]*Grad. Sch. Sci., Univ. Osaka*, [2]*Grad. Sch. Frontier Biosciences., Univ. Osaka*)

Many bacteria swim in aqueous environment by rotating a filamentous organelle, the flagellum. The flagellar axial component proteins are exported through the flagellar type III protein export apparatus to the distal end for self-assembly. The flagellum consists of 20 to 30 thousands of subunits of about 30 different proteins. Thus control of the secretion order and the amount of each flagellar protein is essential to construct such a huge protein complex, but the control mechanism is still unknown. To elucidate the mechanism, we have made up an in-vitro transport assay system using inverted membrane vesicle (IMV). On the basis of the reconstitution experiments of the flagellar axial structure using IMV, we will discuss the regulatory mechanism the secretion order.

3Pos160 リン脂質輸送タンパク質 Sec14 のホスファチジルコリン輸送に対する脂質膜組成の影響
Effect of membrane-components on Sec14-mediated phosphatidylcholine transfer

Taichi Sugiura, Keisuke Ikeda, Minoru Nakano (*Grad. Sch. Med. Pharm. Sci., Univ. Toyama*)

Sec14 is a major phosphatidylinositol (PI)/phosphatidylcholine (PC) transfer protein, which is known to facilitate vesicle budding from the trans-Golgi network in yeast. Sec14 has a hydrophobic pocket and an amphipathic helix that covers a lipid captured into the pocket. However, lipid transfer mechanism of Sec14 has not been fully elucidated. In this study, we evaluated the transfer of pyrene-labeled PC and revealed that PI, phosphatidylserine, and phosphatidylethanolamine facilitate the Sec14-mediated PC transfer. Gel filtration chromatography showed that Sec14 forms oligomer in buffer that dissociates into monomers in the existence of PC or PI. This implies that the state of the protein and its change could control the lipid transfer activity.

3Pos161 線虫（C. elegans）がかぎ分けられるにおい濃度差を行動と神経活動から明らかにする
Specific behavior and neural activity of Caenorhabditis elegans unveil how exactly it detects difference of odor concentration

Hisashi Shidara, Masanari Makino, Kohji Hotta, Kotaro Oka (*Grad. Sch. Sci. and Tech., Keio Univ.*)

C. elegans detects vertical gradients of attractive odorants and gradually curves toward higher concentration to approach odor sources. Although the behavioral strategy, klinotaxis, requires precise detection of the concentration change, it is unknown how exactly worms detect difference of odor concentration. With a microfluidic device to expose a semi-restricted worm to two different laminar flows, we evaluated the odor sensitivities by checking which side worms moved their heads to. In addition, we examined how different concentration of odorants worms could distinguish by visualizing intracellular Ca2+ of olfactory sensory neuron, AWC. These results showed that the estimated sensitivity by Ca2+ imaging was higher than one from the behavior.

3Pos162 視覚ー味覚条件付けによるモノアラガイの視覚特性の検討
Experimental study about the visual characteristics of a pond snail, *Lymnaea stagnalis*, by visual-appetitive conditioning

Satoshi Takigami[1], Momoko Koide[2], Tetsuro Horikoshi[2] ([1]*Grad. Sch. Bio., Tokai Univ.*, [2]*Dept. Biomed. Eng., Sch. Engineering, Tokai Univ.*)

We have investigated the visual characteristics in the *Lymnaea stagnalis*, by using appetitive conditioning. In this study following conditioning paradigm was used: The conditional stimulus (CS) was the presentation of visual pattern of vertical strips, horizontal stripes or check, the unconditional stimulus (US) was the application of sucrose to the lip, and 30 pairs of CS-US presentation for one day. As a result, *Lymnaea* acquired conditioning with the vertical stripes and the check as CS, but not with the horizontal stripes. The animals that conditioned with the check responded also to the vertical strips. These data suggest that the eyes and/or the central nervous system of *Lymnaea* may have a preference mechanism for vertical stripes.

3Pos163 カエル神経筋接合部シナプスでの短期可塑性の二項分布解析: 促通では放出可能な小胞数が増加し、増強では放出確率が増加する

Binomial distribution analysis of short-term plasticity, facilitation and potentiation, at the frog NMJ: n and p increases, respectively

Naoya Suzuki (*Dept. Phys., Sch. Sci., Nagoya Univ.*)

We analyzed short-term synaptic plasticity, facilitation and potentiation, at frog neuromuscular junction using binomial distribution. Facilitation was induced by 8 stimuli with interval of 25 or 30 msec. Potentiation was induced by 500 stimuli at 20 Hz. Then the preparation was kept 5 sec resting to ensure decay of facilitation and the remaining enhancement was maintained as steady state by 350 stimuli with an interval of 150-200 msec. The binominal analyses of the change of **p** and **n** during facilitated and potentiated EPPs distributions showed that increase of **n** and **p** contributed largely to the enhancement of transmitter release in facilitation and potentiation, respectively. The effects of not fixed **p** or **n** were also examined.

3Pos164 培養神経回路網における神経活動パターンの時間遷移の解析

Analysis of the transition of electrical activity patterns in cultured neuronal cells

Takumi Okada, Keisuke Izutani, Hidekatsu Ito, Wataru Minoshima, Suguru N. Kudoh (*Sch. Sci. & Tech., Kwansei Gakuin Univ.*)

The representation of outer world in the brain is undertaken by spatiotemporal activity patterns of neurons. In this study, we attempted to analyze the transition of the internal state of the network. Dissociated rat hippocampal neurons reconstructed a complex network on the particular culture dish with 64 planar microelectrodes on the bottom and spontaneous electrical activity of neuronal network was frequently observed, using extracellular potential multisite recording system. We calculated the center of gravity of the spontaneous activity and analyzed transition of it as the transition of activity patterns. As a result, we confirmed that the limited number of specific patterns repeatedly appeared depending on days in vitro.

3Pos165 やわらかいボディのダイナミクスを用いて情報処理を実装する

Information Processing Using Soft Body Dynamics

Kohei Nakajima[1,2] ([1]*The Hakubi Center for Advanced Research, Kyoto University*, [2]*Graduate School of Informatics, Kyoto University*)

Soft machines have been developed with increasing regularity in recent years. These robots have significant advantages over traditional rigid robots due to adaptive morphology. However, in general, their dynamics are difficult to control due to their intrinsic complexity. In this presentation, we will show that this seemingly undesired property can, in fact, be highly beneficial, in that they can be employed as a computational resource. Using a physical platform consisting of a soft silicone arm, we demonstrate that body dynamics generated by the arm can be used for real-time computations. Our results suggest that the soft body dynamics have a comparable information processing capability with conventional recurrent neural networks.

3Pos166 ランビエ絞輪近傍の BK チャネルは軸索の高頻度発火を制御する

Paranodal BK channels regulate high frequency firing in myelinated axons

Moritoshi Hirono, Hiroaki Misonou (*Grad Sch Brain Sci, Doshisha Univ*)

Voltage-gated potassium channels are uniquely situated at and near the node of Ranvier and thought to play crucial roles in regulating action potentials (APs) in the axon. Recently we obtained results that the calcium- and voltage-activated BK potassium channel is expressed in the paranodal region of myelinated axons in cerebellar Purkinje cells (PCs). To test the role of paranodal BK channels in AP propagation, we examined the failure rate of the antidromic APs upon repetitive stimulation at 50-300 Hz. Local axonal applications of BK channel blockers and nickel significantly increased the failure rate, suggesting that axonal BK channels uniquely support high-fidelity firing of APs in myelinated PC axons, thereby underpinning the output of the cerebellar cortex.

3Pos167 In-situ 光照射固体 NMR による ppR/pHtrII 複合体の光反応過程の解析

Hotoreaction pathway of ppR/pHtrII as revealed by in-situ photo irradiation solid-state NMR

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ppR(pharaonis phoborhodopsin) is a photo receptor with a retinal chromophore, and forms complex with the cognate transducer protein pHtrII to express negative phototaxis through K-, L-, M-, O-intermediates during photocycle. To analyze the retinal configurations during the photocycle, we have developed in-situ photo-irradiation solid-state NMR apparatus to investigate the photo intermediates with the retinal configuration in [14,15-^{13}C]Ret,[ε-^{15}N]Lys-labeled ppR/pHtrII. This NMR system enables to illuminate the multiple LED lights at 520, 595 and 365 nm under a magic angle spinning condition. ^{13}C and ^{15}N NMR signals demonstrated that some intermediates such as the M, O- and N-intermediates like bacteriorhodopsin were stationary observed.

3Pos168 小角 X 線溶液散乱によるアレスチンとロドプシンの相互作用の解析

Interaction between visual arrestin and membrane-embedded rhodopsin studied by solution small-angle X-ray scattering

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The phototransduction cascade in rod visual cell is initiated by the photoactivation of rhodopsin, which enables the activation of retinal G protein transducin. It is terminated by phosphorylation of rhodopsin, followed by the binding of arrestin. Here we analyzed the formation of rhodopsin/arrestin complex by the solution small-angle x-ray scattering in detergent-free system by using rhodopsin embedded in nanodiscs (Rh/ND). Rh/ND was mono-dispersed, and its apparent molecular weight estimated by the intensity of forward scattering agreed with the calculated value (108 KDa). Using this system, the binding of Rh/ND and arrestin (45 kDa) was quantitatively analyzed at physiological concentration, where arrestin self-associates to form tetramer.

3Pos169 シロイヌナズナ phototropin 2 のＸ線小角散乱による構造研究

Small-angle X-ray scattering study of Arabidopsis phototropin 2

Mao Oide[1,2], Koji Okajima[1,2], Yuki Sekiguchi[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Takaaki Hikima[2], Masaki Yamamoto[2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*)

Phototropin (phot), a blue-light (BL) receptor protein of plants, responses to maximize the efficiency of photosynthesis. Phot comprises two light-oxygen-voltage sensing domains (LOV1 and LOV2), that absorb blue light, and a serine/theroine kinase domain (STK) responsible for BL-dependent autophosphorylation leading to cellular signaling cascades. BL-excited LOV2 is primarily responsible to activate STK. But, the molecular mechanism to transmit small conformational changes in LOV2 to STK is still unclear. To understand the mechanism, we investigated full-length of phot2 from Arabidopsis by using small-angle X-ray scattering (SAXS) at SPring-8. The molecular shapes and BL-induced structural changes of phot2 will be discussed in the presentation.

3Pos170 ロドプシンはラフト親和性の短寿命ナノドメインを形成しながら拡散している

Rhodopsin diffuses in disc membranes by forming raftophilic and transient nanodomains

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The high amplification gain of the initial step of phototransduction is explained by the rapid diffusion and collision of photoreceptor rhodopsin (Rh) and its cognate G protein transducin (Gt). However, the diffusivity and even the distribution of these essential components in the disc membranes remain unclear. Here, we analyse single particle tracking data of Rh and Gt in native disk membranes by vbSPT software, which infers the number of diffusive states and transition parameters through variational Bayes treatment of hidden Markov model. Our results demonstrate that both Rh and Gt are diffusing in the disc membranes by making transitions between 3 diffusive sates. Highly raftophilic transient “Rh-cluster-raft” is suggested to present in the disc membranes.

3Pos171 トランスデューシン活性化能を有するロドプシン褪色中間体と Meta II 中間体の同一性に関する検討

Does Meta II correspond to R*, the bleaching intermediate activating transducin?

Shuji Tachibanaki[1,2], Ryota Kumakura[1], Whei-Ee Tang[1], Yoichiro Fukunaga[1], Satoru Kawamura[1,2] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*)

In the vertebrate retina, rhodopsin is the visual pigment in rods. After light absorption, rhodopsin changes its conformation to take an activated state, generally called Rh*, which activates a trimeric G-protein, transducin (Tr). So far, it is widely believed that Meta II, a bleaching intermediate of rhodopsin, is Rh*. In this study, after giving a light flash, by measuring the decay of the activity to activate Tr and by measuring the decay of meta II spectrophotometrically, we compared R* decay and Meta II decay. It was found that Rh* decays faster than Meta II. The result suggests strongly that some specific and transient conformational state during a Meta II period corresponds to R* and is responsible for activation of Tr.

3Pos172 Ab initio MD シミュレーションによる PYP の活性部位水素結合ネットワークの解析

Ab initio MD study on the dynamic structure of the hydrogen bond network in the active site of PYP

Yusuke Kaneta, Hiroshi Watanabe, Tadaomi Furuta, **Minoru Sakurai** (*Center for Biol. Res. & Inform., Tokyo Tech*)

According to neutron diffraction analysis for PYP, there is a low barrier hydrogen bond (LBHB) adjacent to the reaction center: it is
formed between the phenolic oxygen of the chromophore and the carboxylic oxygen of Glu46, In addition, Arg52 is deprotonated. Recently, QM/MM calculations by Saito and Ishikita and electronic structure calculations combined with the solution of the nuclear Schroedinger equation by Nadal-Ferret et al. have provided negative and positive interpretations on these experimental results, respectively. Therefore, the “LBHB problem of PYP” is still controversial. In this study, we address this problem by means of ab initio MD simulations and elucidate the dynamic structure of the hydrogen bond network in the active site.

3Pos173 PYP - Phytochrome Related Protein の発色団が構造へ与える影響

Effects of chromophores of PYP - Phytochrome Related Protein on the structure

Keito Yoshida, Kento Yonezawa, Yoichi Yamazaki, Mikio Kataoka, Hironari Kamikubo (*Grad. Sch. Mat. Sci., NAIST*)

Ppr is a novel light-sensor hisitidine kinase, which comprises of two light sensor domains, PYP and Bph domains. We have revealed that the photo-reactions of PYP domain and Bph domain are interdependent. In order to understand the mutual interdependency, we prepared Apo-Holo-Ppr and Holo-Apo-Ppr, which lack either the chromophore of PYP or that of Bph, and carried out the SAXS measurements. In the results, while the R_g of Apo-Holo-Ppr, 54 Å, is close to that of the intact Ppr, 54 Å, the R_g of Holo-Apo-Ppr is slightly increased to be 58 Å. The increase in R_g of Holo-Apo-Ppr suggests that the alteration in the chromophore of Bph domain would influence the photoreaction of PYP domain through the structural change in the intact Ppr.

3Pos174 液体中ナノ空間拘束下の量子効率増強：色素分子の光学的ホールバーニング過程

Enhancement of quantum efficiency in a nanometer-sized confinement in liquids: optical hole-burning processes of dye molecules

Hiroshi Murakami (*KPSI, JAEA*)

The efficiency of a photochemical reaction in liquids decreases owing to relaxation processes. In biological systems, various strategies have been developed so that the efficiency may not decrease. Diffusional and vibration-like relaxations of water molecules are suppressed at room temperature in small reverse micelles (Murakami, et al. Phys. Rev. E, 2013.). This property is considered to lead to the enhancement of the efficiency of quantum processes in reverse micelles. Thus, in the present study, we examine such an enhancement for the optical hole-burning process of dye molecules in reverse micelles. Water-soluble molecules, including biomolecules, can be dissolved in the nanometer-sized aqueous cavity of the reverse micelles, which could be a model of cells.

3Pos175 Isotope-labeled DNA substrate revealed site-specific interaction with CPD photolyase

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CPD photolyases are flavoproteins that repair CPD-type UV-induced damaged DNA by use of near UV/blue light. Previously, we monitored the photoactivation and DNA repair of E. coli CPD photolyase by light-induced difference FTIR spectroscopy and succeeded in the assignment of some signals [1-2].
To precisely elucidate the FTIR signals of DNA, we developed ^{18}O-labeled CPD substrate that contains C_4=^{16}O and C_4=^{18}O groups at the 5' and 3' side, respectively. We succeeded in distinguishing site-specific vibrations, and the functional role of this site-specific interaction in CPD photolyase will be discussed.
[1] I M. M. Wijaya et al. Biochemistry 52, 1019 (2013).
[2] I M. M. Wijaya et al. Biochemistry 53, 5864 (2014).

3Pos176 Crystallographic study of the LM intermediate of squid rhodopsin

Midori Murakami, Tsutomu Kouyama (*Grad. Sch. Sci., Nagoya Univ.*)

The signal transduction process of squid rhodopsin is through Rhodopsin -> Batho -> Lumi -> LM -> Acid meta to stimulate the Gq protein and is hit back to the initial state. In this study, we performed crystallographic analyses of the LM intermediate of squid rhodopsin.
When the P62 crystal at 100 K was exposed to blue light, an equilibrium state of three isomeric states (rhodopsin, Batho and Iso) was established. When this crystal was warmed to 240K, Batho was converted to LM with no change in the contents of the other states. Diffraction data recorded at 100 K from this crystal were compared with those from the dark-adapted crystal, and the associated difference map indicates that the formation of LM induces conformational changes in the vicinity of retinal.

3Pos177 光合成のマルチ時間スケールダイナミクスに対するシステム解析
Systems approach to the multi-timescale dynamics of photosynthesis

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Photosystem II (PSII) and Photosystem I (PSI) are the protein complexes which drive photosynthesis in tandem employing electron and excitation-energy transfer processes over a wide timescale range from picoseconds to milliseconds. While the fluorescence emitted by the antenna pigments of these complexes is known as an important indicator of the activity of photosynthesis, its interpretation was difficult because of the complexity of reactions in thylakoid membrane. In this study, extensive kinetic models of reactions in thylakoid membrane are analyzed through the use of the hierarchical coarse-graining method proposed in the authors' earlier work. Our novel coarse-grained models of PSII and PSI appropriately describe photosynthetic induction phenomena.

3Pos178 ホウレン草の PSII 反応中心に生成する光電荷分離状態の時間分解 EPR
Time resolved EPR study on photoinduced primary charge-separated state of the PSII reaction center from spinach

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In the initial stage of the photosynthesis in the plants, the photoinduced sequential electron transfers are known to take place in the photosystem II reaction center. It is suggested that the charge-separated (CS) state composed of P(D1) radical cation in the special pair and pheophytin radical anion is generated via the excited singlet state of the accessory chlorophyll. We have employed the time-resolved EPR method and obtained the spectra. The broad spectrum is assigned to the excited triplet state of ChlD1 generated by the triplet charge recombination via S-T0 mixing, the narrow spectrum can be assigned to the initial CS state. We have characterized the orientational structure and the dynamics of at least two kinds of the initial CS states from the narrow signal.

3Pos179 Thermochromatium tepidum 由来 LH1-RC 複合体における耐熱化の分子機構：同位体置換体の FTIR 分析
Molecular mechanisms for the enhanced thermal stability of LH1-RC complex from Thermochromatium tepidum: isotope-edited FTIR spectroscopy

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Molecular mechanisms for the enhanced thermal stability of the light-harvesting 1 reaction center (LH1-RC) complex from Thermochromatium (Tch.) tepidum were investigated by isotope-edited ATR-FTIR spectroscopy. The ATR-FTIR difference spectrum induced by Ca2+-to-Ba2+ substitution exhibited conformational changes of polypeptide backbones and amino acid residues nearby the Ca2+-binding site of LH1-polypeptides. Upon universal 15N- or 13C-labelings, the characteristic Ba2+/Ca2+ difference bands exhibited 15N/14N-, 13C/12C-, or 2H/1H-isotopic shifts but were largely insensitive to 40Ca-labeling. Based on these findings, metal-sensitive FTIR bands were tentatively assigned and the molecular mechanisms responsible for the enhanced thermal stability were discussed.

3Pos180 Formation of complexes between genetically modified photosystem I and single-walled carbon nanotubes

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Carbon nanotubes are new material for their remarkable electronic properties. In this study, complexes between photosystem (PS) I and single-walled carbon nanotubes (SWNTs) were formed by SWNTs binding peptides, which introduced to the reducing site of PS I complex. It expects that the complexes of PS I-SWNT establish efficient electron transfer system. The binding peptide was inserted into the PsaE subunit of PS I in Synechocystis. Because, ferredoxin binding site is close to PsaE. Thus, the SWNT is expected to accept electrons from the Fe-S cluster of PS I. Our results of fluorescence, photoluminescence and electric characteristic measurement clearly showed that the PS I -SWNT complex was formed successfully. We will discuss the properties of the PS I -SWNT complexes.

3Pos181 光合成水分解 Mn_4CaO_5 クラスターの光活性化における Mn^{2+}結合部位の ATR-FTIR 検出

ATR-FTIR detection of the Mn^{2+} binding site in photoactivation of the water-oxidizing Mn_4CaO_5 cluster in photosystem II

Akihiko Sato, Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Univ. Nagoya*)

The Mn_4CaO_5 cluster, which is the catalytic center of photosynthetic water oxidation in photosystem II (PSII), is formed through a light-driven process, called photoactivation. The detailed mechanism of photoactivation remains unresolved. In this study, we detected the initial binding site of Mn^{2+} in the photoactivation process using attenuated total reflection Fourier transform infrared spectroscopy (ATR-FTIR). FTIR difference spectrum upon binding of Mn^{2+} showed two bands both in the symmetric and asymmetric stretching regions of carboxylate groups. The intensities of these bands were virtually unchanged in the range of 20-80 μM Mn^{2+}. These results suggested that two specific carboxylate groups are responsible for the first binding of Mn^{2+} in the PSII protein.

3Pos182 光化学系 II の部位特異的変異導入によるクロロフィル二量体 P680 の電子構造の

Electronic structure of the chlorophyll dimer P680 modified by site-directed mutation at a nearby amino acid residue in photosystem II

Motoki Yamaguchi, Ryo Nagao, Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

Photosystem II (PSII) is a multisubunit pigment-protein complex catalyzing light-driven water oxidation. The reaction center chlorophyll P680, which consists of a pair of chlorophyll molecules, PD1 and PD2, has an extremely high redox potential of about +1.2 V that is necessary for water oxidation. To elucidate the electronic structure of the P680+ cation and its relevance to the high redox potential, we introduced site-directed mutation at D1-V157, which is located near P680, to His using Synechocystis sp. PCC 6803, and studied the effects of mutation on the charge distribution and the redox potential of P680 using FTIR and thermoluminescence. The results showed that the positive charge is mostly located on PD1, and is slightly shifted to the PD2 side in the mutant.

3Pos183 重イオン線照射によってヒト正常線維芽細胞に誘導されるバイスタンダーシグナル

Heavy-ion induced bystander signaling in normal human fibroblasts

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A radiation-induced bystander response is known as a cellular response induced in non-irradiated cells (bystander cells) by receiving bystander factors released from irradiated one. Nitric oxide (NO) is well known as a major initiator/mediator of bystander response. Here, we studied NO-mediated bystander signaling induced by high-linear energy transfer argon-ion microbeam irradiation of normal human fibroblasts. DNA double-strand breaks and phosphorylation of NF-κB and Akt were induced through NO-mediated bystander signaling in both irradiated and bystander cells. However, accumulation of cyclooxygenase-2 was observed in only the bystander cells. Our findings suggest that these signaling molecules play important roles in NO-mediated bystander responses.

3Pos184 超音波によるケージド化合物の活性化のメカニズム

Mechanism of activation of caged compounds induced by ultrasound

Haruko Koura[1], Asuka Kato[1], Masato Mutoh[2], Wakako Hiraoka[1] ([1]*Dept. Phys., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Mater. & Human Env. Sci., Shonan Inst. of Tech.*)

The activity of photoactivatable caged compounds is masked by a photolytic caging moiety. However, photoirradiation may pose a disadvantage in clinical applications. We have investigated the potential of ultrasound as a substitute for photoirradiation. The aim was to determine the mechanism of activation and to determine whether ultrasound-activatable caged compounds have potential for application in medical practice. After activation, the products were analyzed by fluorospectrometry and HPLC. Analysis of the degradation products indicated that the mechanism of release of active moiety by ultrasound irradiation was distinct from that by photoirradiation. Probable mechanisms, such as mechanical stress, radical production, and sonoluminescence, should be considered.

3Pos185 凍結融解を用いたリポソーム融合手法の確立と継続的な RNA 複製反応を伴う人工細胞増殖システムの構築

Reconstitution of artificial cell growth by coupling the RNA replication and propagation in liposomes using a freeze-thawing method

Gakushi Tsuji[1], Satoshi Fujii[2], Takeshi Sunami[3], Tetsuya Yomo[1,2] ([1]*Fbs, Osaka University*, [2]*IST, Osaka University*, [3]*IAI, Osaka University*)

Reconstitution of life-like structure is one of the most challengeable themes for society to elucidate the border between life and non-life. Although many biochemical reactions have been reconstituted inside the artificial lipid-bilayer compartments called liposomes, reactions inside the liposomes were not sustainable and a continuous cell cultivation system had not been reconstructed. In this study, we have developed a novel liposome fusion method by freeze and thawing the reactive liposomes with nutrient liposomes. By using this method, nutrients were supplied repeatedly, enabling the sustainable RNA replication in liposomes. Moreover, liposome membranes were also supplied and subsequently we succeeded to generate the daughter liposomes containing the replicated RNA.

3Pos186 大腸菌クローン集団の抗生物質に対する不均一な応答

Heterogeneous Responses to Antibiotic stress in a Clonal Population of *Escherichia coli*

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Phenotypic plasticity is the ability of cells to commit favorable phenotypic changes without mutations in response to environmental conditions, which is suggested to contribute to the adaptive evolution of antibiotic resistance. However, how phenotypic plasticity contributes to the adaptive evolution remains unclear. Here, we explored responses of *E.coli* to kanamycin (KM) stress at the single cell level. Interestingly, KM treatment induced heterogeneous responses in a clonal population, i.e., some cells died while the other cells continued to divide. The results suggested that the cellular fate depended on the ancestral cells' states several generations before KM treatment. Based on the results, we will discuss the role of phenotypic plasticity in adaptive evolution.

3Pos187 細胞外刺激前後のクロマチンダイナミクスの変化の 1 分子イメージング解析

Single molecule imaging analysis of chromatin dynamics in response to extracellular stimulation

Kayo Hibino[1,2], Ryosuke Nagashima[1,2], Kazuhiro Maeshima[1,2] ([1]*NIG*, [2]*SOKENDAI*)

Chromatin dynamics, such as local fluctuation and long range movement, are involved in DNA replication and transcription, which specify cell fates. These DNA transactions also have influence on chromatin dynamics themselves mutually. However, fundamental information on chromatin dynamics is still unclear, especially its response to extracellular stimulation. Here, we investigate chromatin dynamics before and after extracellular stimulation in living cells. Single nucleosome movements were observed in normal epithelial RPE1 cells by using single molecule imaging techniques. In the meeting, we will discuss responses of chromatin to various stimulations.

3Pos188 タンパク質における分子トンネルの形状分析

A Method for Detecting and Analyzing Shapes of Molecular Tunnels in Proteins

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Protein realizes its function by its particular shape such as groove, cleft, pocket and tunnel. It has been reported that molecular tunnels are involved in transferring substrates in reaction processes, but a molecular characteristic of the tunnel remains elucidated. Here we develop a simple method based on grid coordinate that searches the proteins for a tunnel and applied this method to several proteins that are known to have a tunnel. We found that the shapes of the tunnels were variable such as curved and partially squeezed by the irregular lining structure of the tunnel. Even in the same protein crystallized in difference conditions, the shapes of the tunnels were significantly different. By our new method, the molecular nature of the tunnels can be described.

3Pos189 剛体アンサンブルドッキングによって得られた候補構造群における相互作用残基ペアの特徴の解析

Analysis of background interaction properties of protein complexes generated by rigid-body ensemble docking

Nobuyuki Uchikoga[1], Yuri Matsuzaki[2], Masahito Ohue[3], Yutaka Akiyama[2,3] ([1]*Dept. of Physics, Chuo Univ.*, [2]*ACLS, Tokyo Tech*, [3]*Grad. Sch. of Inform. Sci. and Eng, Tokyo Tech*)

We used rigid-body docking algorithm generating many possible protein complexes, called decoys. To understand mechanisms of protein-protein interactions, we focused on some physicochemical properties, electrostatic and hydrophobicity, of a set of docking decoys, generated by a rigid-body docking process. From these decoys, we obtained sets of possible interacting amino acid pairs, involved in background interaction properties. Ensemble docking process by MEGADOCK ver. 4.0 and ZDOCK ver. 3.0.1 is used for analyzing background properties, using datasets composed of typical 44 protein pairs and of 8 proteins involved in MyD88 pathway. In this work, we investigated mainly differences between true and false protein pairs.

3Pos190 アミノ酸プロファイルによるタンパク質ペプチド複合体のポストドッキング解析

Post-docking analysis of protein-peptide complex structure using amino acid profiles

Masahito Ohue[1], Nobuyuki Uchikoga[2], Yuri Matsuzaki[3], Yutaka Akiyama[1,3] ([1]*Dept. Comput. Sci., Grad. Sch. Inform. Sci. Eng., Tokyo Tech.*, [2]*Dept. Phys., Facul. Sci. Eng., Chuo Univ.*, [3]*ACLS, Tokyo Tech.*)

Protein-peptide interactions, where one partner is a globular protein and the other is a flexible linear peptide, are important for understanding cellular processes and regulatory pathways, hence they are targets for drug discovery. In this study, we applied rigid-body protein-protein docking software to protein-peptide complexes with multiple peptide conformations. Then we analyzed amino acid profiles for all peptide conformations and docking poses.

3Pos191 タンパク質構造の持つトポロジーの安定性を評価する

How to estimate topological stability of protein structures

Shintaro Minami[1], George Chikenji[2], Motonori Ota[1] ([1]*Dept. of Comp. Sci., Nagoya Univ.*, [2]*Dept. of Eng., Nagoya Univ.*)

Protein fold can be characterized by the spatial arrangement of Secondary Structure Elements (SSEs), and the order of SSEs along the protein sequence (connectivity). It is widely accepted that the total number of stable protein folds is limited by physicochemical rules. To understand the rules, we previously analyzed a limitation of spatial arrangement of SSEs comprehensively. In this study, we analyzed the connectivity observed in the current PDB structures using non-sequential structure alignment program MICAN. Based on the statistics and theoretical calculations, we estimated instability of rare connectivity features such as "loop crossing" or "left-handed β-α-β motif". We will discuss the rules of connectivity, designability, and propose designable new folds.

3Pos192 β-Trefoil タンパクのフォールディングコアの残基間平均距離統計に基づく予測

Prediction of folding nuclei of beta-Trefoil proteins based on the inter-residue average distance statistics

Takuya Kirioka, Norihiro Kanemaru, Takeshi Kikuchi (*Ritsumeikan University*)

The β-Trefoil fold has pseudo threefold symmetry and three repeating subdomains. The relationship between folding pathway and the amino acid sequence of a β-Trefoil protein is very interesting. In this study, we try to predict significant regions of the 3D structure of a β-Trefoil protein formation in its amino acid sequence from the prediction of compact region by contact map based on inter-residue average distance statistics (Average Distance Map), and the prediction of high interacting residues (F value analysis). Furthermore, we examine conserved hydrophobic residues, and try to identify significant residues for folding of the β-Trefoil architecture from an amino acid sequence.

3Pos193 サポートベクターマシンを用いたアミノ酸配列からの脂質結合予測および脂質結合残基予測

Predicting lipid-binding proteins and lipid-binding residues from amino acid sequences by using support vector machine

Kokoro Ueki, Shugo Nakamura, Kentaro Shimizu (*Grad Sch. of Agr., The Univ. of Tokyo*)

Lipid binding proteins (LBPs) have various essential functions in organisms. In this study, we developed methods using support vector machine for predicting LBPs and lipid-binding residues from their sequences. LBPs and non-LBPs were collected from Uniprot for the prediction of LBPs. On the other hand, lipid-binding residues and non-lipid-binding residues were collected from LBPs at the Protein Data Bank for the binding residue prediction. We considered position specific scoring matrix and several predicted physicochemical properties as features that characterize LBPs and lipid-binding residues. Prediction performances were evaluated by 5-fold cross-validation. Our prediction methods for LBPs and lipid-binding residues outperformed the methods proposed previously.

3Pos194 少数個体がつくる鮎の群れに内在する相互作用構造

Interaction Structure of fish schools in a small population

Takayuki Niizato[1], Hisashi Murakami[2], Kazuki Sangu[1], Yuta Nishiyama[3], Kohei Sonoda[4], Yukio Gunji[2] ([1]*Tsukuba University*, [2]*Waseda University*, [3]*Osaka University*, [4]*Shiga University*)

We investigate a fish schooling behavior in a small population. Ayu (Plecoglossus altivelis) makes a group when they are in an unfamiliar environment. In this study, we never confirmed many self-similar structures in a given time series, but also found specific types of interaction structure in a fish school. We suppose that our finding about these interaction structures have a relationship as figure-ground in a fish school.

3Pos195 マイクロアレイデータに基づく植物の遺伝子発現揺らぎと機能の関係

Analysis of relationship between noise of gene expression and function of plants based on microarray data

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Various functions of the organism are realized through appropriate gene expressions. The gene expression level has stochastic variations among genetically identical organisms in the same environmental conditions. (called gene expression “noise”)
Recently, experiments of yeast and Escherichia coli suggested that Plasticity (the difference of the gene expression depending on an environmental change) and Noise of gene expressions in usual environment have positive correlation like the Fluctuation-dissipation theorem. In addition correlation between gene fluctuation and response depend on importance and promoter, gene expression levels is suggested, too.
Therefore we researched that on Arabidopsis thaliana using plural public databases from AtGenExpress.

3Pos196 3 次元細胞シミュレーションにおける高次精度・高解像度スキームの評価

Evaluation of higher order and high resolution schemes for 3D cell simulation

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3D cell simulator (3DCS) is for computing spatial distributions of quantities of biological substances in time inside cells. Although such as VCell is publicly available, currently precise whole 3DCS is not expected. We are developing a novel 3DCS aimed at first implementation of equations of motion in 3DCS, which is expected in future synthetic biology, etc., as like as computational fluid dynamics in aerospace and automotive engineering. It has implemented the central difference (CD) and modified WENO schemes (mWENO). We computed quantities of two hypothetical substances initially distributed as data reconstructed from 68 confocal laser microscopic images of a BY2 cultured cell. They resulted in smooth distributions in mWENO whereas fatal numerical oscillations in CD.

3Pos197 分子の種類を識別する細胞情報処理の確率モデルとその情報論的考察

Stochastic modeling and information-theoretical study of molecular discrimination

Masashi K. Kajita[1], Kazuyuki Aihara[1,2], Tetsuya J. Kobayashi[2] ([1]*Grad. Sch. Inf. Sci. Tech., Univ. Tokyo*, [2]*IIS, Univ. Tokyo*)

In cellular information processing systems, species of molecules are used as information carriers. For accurate information processing, the systems have to discriminate similar molecules to avoid crosstalk among the different carriers. Recent single-cell-imaging data suggest the importance of adaptive phenomena (i.e., receptor clustering) in the cellular information processing. In this research, we use a stochastic model of T cell antigen discrimination, and discuss the possible function of the adaptive kinetics from the viewpoint of information theory.

3Pos198 On thermodynamics of macromolecular association in vivo: Theoretical and simulation studies with a coarse-grained model

Tadashi Ando[1], Yuji Sugita[1,2,3,4], Michael Feig[5] ([1]*RIKEN QBiC*, [2]*RIKEN AICS*, [3]*RIKEN iTHES*, [4]*RIKEN TMS*, [5]*Michigan State Univ.*)

The macromolecular crowding alters the thermodynamics and kinetics of biological reactions and processes in living cells. Here, to broaden our view of in vivo crowding effects, we have evaluated the stabilization on macromolecular association due to volume exclusion of crowding by using both simulations and a theory with a coarse-grained model of a bacterial cytoplasm. Degrees of stabilization evaluated by the simulations and theory were in good agreement each other. For ordinary proteins, the stabilization in the modeled cytoplasmic space was less than -3 k_BT. The present study using the simple model would provide some controls for further studies on in vivo crowding.

3Pos199 Clausius Inequality in Population Growth

Yuki Sugiyama, Tetsuya J. Kobayashi (*IIS, Univ. Tokyo*)

Steady state thermodynamics (SST) was established for understanding a "physics" of transitions between nonequilibrium steady states (NESS). In this theory, by dividing heat into housekeeping part (which maintains NESS) and excess part (which makes relaxation to NESS), one reformulates Clausius inequality in nonequilibrium situations.
In this presentation, we find SST framework in population dynamics. According to SST methods, we divide population growth in fluctuated environment into housekeeping and excess parts. Then, focusing on latter, we obtain Clausius inequality giving the upper bound of excess growth and find that this bound is evaluated by using "lineage fitness". Furthermore, we prove that this upper bound is archived in quasistatic environmental changes.

3Pos200 バクテリアケモスタットのためのマイクロ流体デバイスの構築

Development of microfluidic device for bacterial chemostat

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A chemostat is a continuous cultivation system for cells, which allows us to maintain cell density and condition for a long time; the chemostat is therefore used for many fields such as metabolic/genetic engineering. Especially, micrometer-sized chemostats for single-cell observation have recently been required for the use in synthetic biology. In this study, we propose a microfluidic chemostat based on water-in-oil microdroplets. The rate of continuous flow of medium was controlled by the frequency of droplet fusion and fission. We numerically predicted suitable experimental conditions, and are constructing the microhemostat in a microfluidic device. We believe that this chemostat will be applied to a wide range of studies in synthetic biology.

3Pos201 匂いを嗅ぎ走り出す走化性液滴：ガス刺激からの逃避行動への応答

Chemotactic behavior of a liquid droplet: Smelling and escaping against evil gas

Hiroki Sakuta[1], Nobuyuki Magome[2], Yoshihito Mori[3], Kenichi Yoshikawa[1] ([1]*Facul. Lif. Med. Sci., Doshisha Univ.*, [2]*Chem., Dokkyo Med. Univ.*, [3]*Facul. Sci., Ochanomizu Univ.*)

Living organisms on the earth exhibit self-propelled motion in response to outer stimuli, chemotaxis, where the motion is driven by chemical energy under isothermal condition. We will report an artificial model mimicking the chemotactic behavior of living organism, by adapting an experimental system of an oil droplet floating on aqueous layer. We observed the escaping motion of cm-sized droplet of oleic acid in response to ammonium vapor. After administration of ammonium, the droplet began to escape from the stimulating gas, continued the motion for the length of several cm. The mechanism such chemotactic behavior will be discussed in terms of the induction of the spatial gradient on interfacial tension through the action of the stimulating gas.

3Pos202 In situ マイクロ波照射 NMR 法を用いた生体系におけるマイクロ波効果の解析

Analysis of microwave effects on biological system by in situ microwave irradiation NMR spectroscopy

Yugo Tasei[1], Takuya Yamakami[2], Fumikazu Tanigawa[1], Izuru Kawamura[1], Teruaki Fujito[3], Kiminori Ushida[2], Motoyasu Sato[4], Akira Naito[1] ([1]*Graduate School of Engineering, Yokohama National University*, [2]*Department of Chemistry, School of Science, Kitasato University*, [3]*Probe Laboratory Inc.*, [4]*Faculty of Engineering, Chubu University*)

Microwave heating is widely used to accelerate organic, polymerization and enzymatic reactions. However, the detailed microwave effect on chemical reactions is not yet fully understood. We have developed an in situ microwave irradiation NMR spectroscopy to investigate the effects of microwave heating on organic compounds. The temperature of sample under microwave irradiation was determined using temperature dependent chemical shifts of individual protons in the molecule. As a result, the protons in the polar group exhibited significantly different temperatures in the molecule. These results suggest that microwave heating induce polarization in polar groups, and these effects may occur considerably in biological systems.

3Pos203 光制御水素化アモルファスシリコン薄膜上の化学反応性積層ゲルを用いた植物由来分子の検出

Detection of plant derived molecules using electrochemical laminated gels photo-controlled on hydrogenated amorphous silicon film

Shotaro Minato[1], Yutaka Tsujiuchi[1], Hiroshi Masumoto[2], Takashi Goto[3] ([1]*Material Science and Engineering, Akita University*, [2]*Frontier Research Institute for Interdisciplinary Sciences, Tohoku University*, [3]*Institute for Materials Research, Tohoku University*)

On an attempt for fabrication of biosensor using photo-controlled film system, we have been researching, by using ionic conduction in laminated gels on hydrogenated amorphous silicon film. Ionic conduction in solution as well as in intermediate states between a liquid and a solid, such as gels, is a key phenomenon to inter-conversion between light energy, chemical energy, and electric energy, are able to be achieved using amino acids that is the elementally element of bio molecule and has potential of diversity to electro chemical device. In this conference, we report an attempt of detecting plant derived molecules.

3Pos204 バクテリアプロトプラストとマイクロ膜チャンバーの融合細胞内部の ATP 濃度の測定

Measurement of ATP concentration in fusion cell of bacterial protoplast into micro-sized lipid membrane chamber

Hiroki Ashikawa[1], Kazuhito Tabata V.[1,2], Rikiya Watanabe[1,3], Hideyuki Yaginuma[3,4], Hiroyuki Noji[1,3] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*PRESTO, JST*, [3]*CREST, JST*, [4]*QBiC, RIKEN*)

We have been trying to develop artificial cell system by fusing a protoplast of E.coli to a micro chamber sealed with lipid bilayer and succeeded in observing the fusion. However, there is no established method to measure the inner state of the fusion cells. In this study, we developed methods of measuring ATP concentration ([ATP]) in the fusion cells with BQueen (ratiometric fluorescent ATP indicator) and measured [ATP] and time course. As an observed example, although [ATP] in the cell just after fusion was as high as before, after 3 hours [ATP] declined less than limit of detection. This suggests fusion cells consume ATP more rapidly than they synthesize. In the future, with these methods, we explore the inner state in which ATP is more synthesized.

3Pos205 超高感度 ELISA の開発：血中 HIV-1 p24 ならびに尿中アディポネクチンの測定

Ultrasensitive ELISA: Detection of HIV-1 p24 in blood and that of adiponectin in urine at attomole level

Etsuro Ito (*Kagawa Sch. Pharmaceu. Sci., Tokushim Bunri Univ.*)

To reduce the window period after infection and to minimize the suffering of patients by a noninvasive test, an increase in sensitivity for the target proteins is needed in clinical tests. For this purpose, we developed an ultrasensitive ELISA coupled with a thio-NAD cycling and applied it to measure HIV-1 p24 in blood and adiponectin in urine. The limit of detection for p24 in blood was ca. 10^{-18} moles/assay, and that for adiponectin in urine was ca. 10^{-19} moles/assay. These values are more sensitive than the ELISA data reported so far. Additional recovery tests using blood and urine supported the reliability of our ultrasensitive ELISA. These results showed that our ultrasensitive ELISA is useful in the diagnosis and treatment of various diseases.

3Pos206 原子間力顕微鏡による力学的単一細胞診断：細胞力学量のばらつきの空間依存性

Single cell mechanical diagnostics using atomic force microscopy: how cell-to-cell mechanical variation depends on the cell position

Maki Sawano[1], Kaori Shigetomi(Kuribayashi)[1], Kinho Shu[1], Ryosuke Takahashi[1], Agusu Subagyo[2], Kazuhisa Sueoka[2], Ryosuke Tanaka[1], Takaharu Okajima[1] ([1]*Grad. Sch. bio., Univ. Hokkaido*, [2]*Grad. Sch. Nano., Univ. Hokkaido*)

Atomic force microscopy (AFM) is a useful tool for a mechanical diagnosis of single cell disease [1]. To realize a reliable mechanical diagnosis using AFM, it is essential to control cell conditions for reducing the intrinsic variation involved in the same cell type. Moreover, the measurement location should be optimized for minimizing the cell-to-cell variability measured in the same cell type. To defining these conditions, we investigated the spatial distribution and variation in rheological property of cells cultured on micropatterned substrates by AFM. We observed the variation in cell elastic modulus decreased around the cell center compared with the cell edge, implying the center is suitable for diagnosing single cells. 1) Cross SE et al. Nat Nanotech. 2(2007)780.

3Pos207 Development of target-specific single-dot chemical probes in live cells via intracellular click reaction

Yanyan Hou[1], Satoshi Arai[1], Tetsuya Kitaguchi[1,2], Madoka Suzuki[1,2] ([1]*WASEDA Biosci Res Inst Singapore (WABIOS)*, [2]*Org Univ Res Initiatives, Waseda Univ*)

Organic dyes have been widely used as chemical probes in live cells. However, specific and stable organelle targeting with a function of sensing cellular environments is still challenging. Here we developed single-dot chemical probes where four organic dyes were conjugated to tetravalent single-chain avidin (scAVD) protein by copper-free click reaction. This probe exhibited increased brightness and photostability compared to its single organic dye counterpart. By changing the targeting-signal fused to scAVD, this probe localized to different cellular compartments. Moreover, by conjugating thermosenstive TAMRA dyes, we generated temperature-sensitive probes monitoring cellular temperature changes in both cytosol and on ER membrane in response to external heat sources.

3Pos208 一分子観察のための高速 AFM /チップ増強蛍光顕微鏡複合機

Combined system of HS-AFM and tip-enhanced fluorescence microscopy for single molecular imaging

Shingo Fukuda[1], Takayuki Uchihashi[1,2], Ryota Iino[3], Toshio ANdo[1,2] ([1]*Sch. Math. & Phys., Col. Sci. & Eng., Kanazawa Univ.*, [2]*Bio-AFM FRC, Inst. Sci. & Eng., Kanazawa Univ*, [3]*Okazaki Inst. Integr. Biosci., NINS*)

Recently, we have developed combined HS-AFM and TIRFM system which can simultaneously capture conformational dynamics of protein and binding/dissociation events of fluorescent ligand at the single-molecule level. However, applicable concentration of fluorescent ligand is limited to nanomolar level due to high background fluorescence. To overcome this issue, we utilized surface plasmon resonance (SPR) which enhances local fluorescence at the vicinity of metallic nanostructure. To induce the SPR at the imaging area of AFM, the silver coated cantilever tip was manufactured. We demonstrate simultaneous HS-AFM / TIRFM imaging of single Cy3-labeled chitinase moving on chitin fibril in the presence of relatively high concentration of Cy3-chitinase.

3Pos209 仮想電子顕微鏡 VEM の開発

Development of virtual electron microscopy to supply unified user interface

Ayaka Iwasaki (*Kyushu Institute of Technology*)

When researchers perform structure analysis with an electron microscope (EM), they control EM through each of user interfaces (UIs), which each EM developer provides. What they have to do is different due to difference of each of UIs, even though the same control of EM is operated. To solve this problem, we have developed novel web client software called WITs providing a unified UI between many EMs, which needs a virtual EM (VEM) to wrap the difference of each EM control, as well. Thus we have developed VEM system. Based on the information of the devices, we wrapped the differences in the operation of each EM, translated them into our proposed interfaces and provided the same interface through VEM. Here we will report the progress of the VEM development.

3Pos210 大量データ計測技術を用いたシアノバクテリアの低温コヒーレントX線回折イメージング

Cryogenic coherent X-ray diffraction imaging of cyanobacteria using the high-throughput measurement techniques

Amane Kobayashi[1,2], Yuki Sekiguchi[1,2], Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2], Yayoi Inui[3], Sachihiro Matsunaga[3], Yuki Takayama[2], Masaki Yamamoto[2] ([1]*Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*, [3]*Sci. Tech., Tokyo Univ. Sci.*)

In coherent X-ray diffraction imaging (CXDI), electron density maps of non-crystalline particles are reconstructed by applying the phase-retrieval method to the Fraunhofer diffraction patterns. Recent years, we have conducted CXDI experiments in the X-ray Free electron laser (XFEL) facility, SACLA. By utilizing the XFEL sources, which provide very intense pulses with repetition period of 10-30 Hz, and the cryogenic diffraction apparatus with high-throughput measurement techniques, we have succeeded to collect a huge amount of diffraction patterns within a few hours. Here, we introduce the measurement techniques and the performance of the apparatus. In addition, we report the preliminary results regarding internal structure of cyanobacteria.

3Pos211 細胞内リン酸化タンパク質の多並列超迅速解析法
Multi-parallel super rapid analysis methods of intracellular phosphoproteins

Naoki Takeuchi[1], Miho Suzuki[1], Akira Kurisaki[2], Koichi Nishigaki[1] ([1]*Grad. Sch. of Sci. and Eng., Saitama Univ.*, [2]*AIST*)

A large amount of experiments are needed in current R & D, leading to the requirement of high performance. In this stream, a novel concept microarray MMV (microarray with manageable volumes: Sharma et al. BMC Biotech. 2014) combined with a highly sensitive detection system: 4SR (Stacked Slice-gel System for Separation and Reactions) +Western blot-like monitoring was developed. Using this, we could perform the monitoring of phosphorylation of proteins within a small number of cells in high parallelism of 100 (or 1024). We applied it to detection of pSmad2 and CREB in CHO cells. In this system, we could attain a high performance P defined below: P=（Output×Quality）/（Time elapsed× Cost). That is ~100 fold higher than the conventional approaches.

3Pos212 シグナル分子の時空間的に異なる局在が走化性を調節している
Spatiotemporal different localizations of multiple signaling molecules mediate chemotaxis in Dictyostelium cells

Yuki Tanabe[1,2], Masahiro Ueda[1,2] ([1]*Graduate school of Science, Osaka University*, [2]*QBiC, RIKEN*)

Chemotaxis, the directed migration in chemical gradients, is a vital function in many biological processes such as immune response and synapse formation. Chemotaxis is mediated by some parallel signaling pathways, e.g. PI3K-path, sGC-path and so on. However, what characteristics each pathway has remains unknown. Here, we observed membrane localization of signaling molecules (PIP3 and sGC) simultaneously and analyzed localization dynamics.
As a result, sGC showed more frequently localization in the absence of chemoattractant. In the presence of chemoattractant, on the other hand, both PIP3 and sGC localized in the same region and cells quickly moved towards chemoattractant. We discuss that each chemotactic signaling pathway has different roles.

3Pos213 CLIP-170 phosphorylation mediates repositioning of microtubule-organizing center during T cell activation

Wei Ming Lim, Yuma Ito, Makio Tokunaga, Kumiko Sakata-Sogawa (*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*)

During initial response of T cell activation, immunological synapse (IS) is formed at the interface between T cell and antigen presenting cell (APC). Microtubule-organizing center (MTOC) is translocated toward the center of IS and drives directional transport of effector vesicles toward bound APC. However, the molecular mechanism of MTOC repositioning remains unclear. Recent studies reported adenosine monophosphate kinase (AMPK) mediates microtubule dynamics by regulating phosphorylation of cytoplasmic linker protein 170 (CLIP-170). CLIP-170 binds to GTP-tubulin with high affinity at the microtubule plus end, while loses affinity upon phosphorylation. Here, we studied the importance of CLIP-170 toward MTOC repositioning by fluorescence quantitative imaging analysis.

3Pos214 リポ多糖刺激における炎症抑制タンパク質 PDLIM2 活性化のイメージング定量解析
Quantitative imaging analysis of anti-inflammatory protein PDLIM2 activation upon LPS stimulation

Shota Ichikawa[1], Yuma Ito[1], Takashi Tanaka[2], Makio Tokunaga[1], Kumiko Sakata-Sogawa[1] ([1]*Grad. Sch. Biosci. Biotech., Tokyo Inst. Tech.*, [2]*IMS-RCAI, RIKEN*)

Activation of NF-κB plays a critical role in the regulation of immune responses. PDLIM2 is an E3 ubiquitin ligase, which terminates NF-κB activation in the nucleus to prevent excessive inflammatory responses. However detailed mechanism of PDLIM2 function is still elusive. Aiming to clarify the regulation mechanism of PDLIM2 by imaging analysis, we established dual gene expressing 3T3 cell lines of GFP- or RFP-fusion proteins of PDLIM2 and NF-κB (p65 subunit). We observed the nuclear translocation of PDLIM2 together with NF-κB upon LPS stimulation. We will discuss the different response of PDLIM2 depending on stimulants.

3Pos215 温度上昇に伴うマクロファージ運動の活性化
Activation of motility of macrophage at temperature jump

Hideo Saitou, Masamine Shintani, Sayaka Kita, Hideo Higuchi (*The university of tokyo*)

Macrophage was activated and performs an immune reaction after alien substances such as bacteria and viruses invaded in our bodies. It is known that the activity of macrophage depends on the temperature. To understand the transient processes of macrophage at temperature jump, we observed the motility of macrophage and vesicle transport. Macrophage was purified from mouse abdominal cavity. The temperature jump was initiated by local illumination of macrophage medium by the infrared laser (λ=1550nm). The macrophage moved toward the heat source while stretching filopodia and lamellipodia with delay of time. The velocity of vesicles labeled with quantum dots (QD655) was accelerated by the heating.

3Pos216 高感度および高解像度電気化学チップデバイスによる幹細胞の機能イメージング
Electrochemical imaging of stem cell function using high-sensitivity and high-resolution electrochemical chip devices

Yusuke Kanno[1], Kosuke Ino[1], Hitoshi Shiku[1], Tomokazu Matsue[1,2] ([1]*Graduate School of Environmental Studies, Tohoku Univ.*, [2]*WPI-Advanced Institute for Materials Research, Tohoku Univ.*)

Microelectrode array chip devices have been developed to obtain high temporal resolution toward bioimaging. Previously, we developed a novel electrochemical device with comb-type interdigitated array (IDA) electrodes, which introduced a local redox cycling-based electrochemical (LRC-EC) system for multi-electrochemical detection and signal amplification. However, it was difficult to densify the sensor points and rise sensitivity because IDA electrodes were fabricated two-dimensionally. In the present study, a three-dimensional configuration of electrodes has been adopted to achieve higher density and sensitivity of electrochemical devices. Furthermore, fabricated devices were applied to electrochemical imaging of the function of mouse embryonic stem cells.

3Pos217 ゼブラフィッシュグリア細胞活動の In vivo イメージング
In vivo imaging of glial cell activity in zebrafish

Hiroko Bannai, Masashi Tanimoto, Shigeo Sakuragi, Yurie Matsutani, Yoichi Oda (*Grad. Sch. Biol. Sci., Univ. Nagoya*)

Glial cells are major component of the central nervous system (CNS), along with neurons. Ca^{2+} signals in glial cells influence synaptic transmission and local blood flow which are essential for brain function in mammalian brain. Althoufh glial cells are also found in non-mammalian animals, whether glial Ca^{2+} signals are involved in the regulation of brain function remains to be elucidated. To examine Ca^{2+} signal in non-mammal vertebrate, we created transgenic zebrafish expressing membrane-targeted genetically encoded Ca^{2+} indicator GCaMP6F under glia specific promoter. In vivo imaging of transgenic zebrafish larvae under spinning disc confocal microscope revealed the existence of local, spontaneous Ca^{2+} signals in glial cells in multiple regions of CNS.

3Pos218 VSFG 検出赤外超解像顕微分光法による羽毛 β-ケラチンの分子配向赤外イメージング
Orientation-sensitive IR imaging of feather β-keratins by a VSFG-detected IR super-resolution micro-spectroscopy

Yukihisa Watase, Kohei Ushio, Masaaki Fujii, Makoto Sakai (*Chem. Res. Lab, Tokyo. Tech*)

Feather is generally known to consist of rachis, barb and barbule regions from the root to the tip, and it has been reported that main components of feather are keratin proteins with β-sheet structures (β-keratins). On the other hand, the spatial inhomogeneity of β-keratins, such as the distribution and orientation, could not be disclosed because of a lack of the spatial resolution of previous analytical methods. In this study, we aim to elucidate the spatial distribution and orientation of β-keratins at each region of feather in the amide I band and verify those differences at each region by a VSFG-detected IR super-resolution microscopy. In the presentation, the orientation of β-keratins will be also discussed with the results of polarization dependent VSFG imaging.

3Pos219 マイクロ波照射下での酵素反応の出力依存性に関する研究
The study of microwave output dependence on enzymatic reaction under microwave irradiation

Fujiko Aoki[1], Kenshi Haraguchi[2], Arata Shiraishi[1], Syokichi Ohuchi[1] ([1]*Dept. Lifesci. & Syst. Eng., Kyushu Inst. Tech.*, [2]*Dept. Biosci. & Bioinform., Kyushu Inst. Tech.*)

In this study, we intended to reveal the microwave output dependence in enzymatic reaction. For example, we examined the hydrolysis reaction of p-nitrophenyl acetate using lipase from Candida rugosa. By using double jacketed reactor for cooling, we controlled the reaction temperature and microwave output exactly. Then, we measured p-nitrophenol formed in 42℃, 45℃ at 0 ～ 100 W and analyzed kinetic parameters. As a result, we turned out that the michaelis constant（Km）and the maximum initial velocity（Vmax）of enzymatic reaction vary for microwave output. In addition, we found that the microwave output dependence of kinetic parameters changes in the reaction temperature.

3Pos220 弾性率可変マイクロファイバーゲルマトリックスにおけるがん細胞の三次元運動表現型評価
Phenotypic differences in 3D movement of tumor cells observed in the microfiber gel matrices with tunable elasticity

Yu Nakamura, Satoru Kidoaki (*Institute for Materials Chemistry and Engineering, Kyushu Univ.*)

Cell motility in vivo is essentially influenced by the mechanical properties such as stiffness of 3D extracellular matrix. To elucidate the mechanistic aspects of the 3D cell motility, systematic design of mechanical conditions of 3D cell culture matrix is quite important. So far we have developed the extracellular-matrix-mimicking elasticity-tunable microfiber gel matrix based on the photo-induced gelation of electrospun nano/microfibers of photocurable gelatin. In this study, metastatic cancer cell (MDA-MB-231) were cultured in the microfiber gel matrices with different elasticity, and compared with the behaviors of normal epithelial cell (MCF-10A). Phenotypic responses of 3D movement of the tumor cell to the mechanical milieu were systematically evaluated.

3Pos221 cDNA display 法によるリポソーム結合ペプチドの試験管内進化
In Vitro Selection of Liposome Anchoring Peptide by cDNA display

Naoto Nemoto, Ryouya Okawa, Yuki Yoshikawa, Toshiki Miyajima, Shota Kobayasi (*Grad. Sch. of Sci. and Eng., Saitama University*)

Liposome-anchoring peptides (LA peptides) were selected from a random peptide (30-residue-length) library by in vitro selection using cDNA display. After six rounds of selection, the random sequences converged to a unique consensus amino acid sequence. The selected peptides were synthesized and modified by the addition of fluorescein at the N- or C-terminus by chemical-synthesis, and examined the interaction of the peptides with liposome membranes using confocal laser scanning microscopy. The LA peptides were interacted with liposomes composed of dioleoyl-sn-glycero-3-phosphocholine (DOPC) over the concentration range of 0.1-6 μM. We will discuss the function of LA peptide and its application.

3Pos222 Clogging of DNA driven through a nano-scale pore or slit

Naoto Sakashita, Yuta Kato, Yoshitaka Tanida, Kentaro Ishida, Toshiyuki Mitsui (*Coll. of Sci. & Eng., Aoyama Gakuin Univ.*)

Nanopore-based sensing technology has revealed the capability of single polymer sensing by measuring the time course of the ionic current through a nanopore. This sensing provides the information of the molecule length and configurations. However, the nanopore sensing has a critical issue that DNA molecules often clog into the pore. To elucidate the clogging mechanism, we have investigated the interactions between a pore and single DNA molecules before their translocations by optical microscopy. We have also measured the probabilities of the clogging vs. pore shapes, such as slit and circle, or DNA length. In this presentation, we will discuss the influence of the knot formation of polymer on the clogging probability and the ideal pore for the reliable nanopore devices.

3Pos223 明視野/蛍光マルチイメージングフローサイトメーターを用いた形状情報認識を用いた細胞識別技術の検討

Studies on identification of cells using visible morphological information using bright field/fluorescent multi-imaging flow cytometer

Akihiro Hattori[1], Hyonchol Kim[1], Hideyuki Terazono[2], Masao Odaka[1], Kenji Matsuura[1], Mathias Girault[1], Kenji Yasuda[1,2] ([1]*Kanagawa Academy of Science and Technology*, [2]*Tokyo Medical and Dental University*)

We have examined the ability of real-time identification of cells by simultaneous measurement of bright field/fluorescent images of cells in an on-chip multi-imaging flow cytometer system. First, we measured the depth dependence of images and blurs of cells in the microfluidic flows by introducing the fluorescent polystyrene beads and HeLa cells stained with Hoechst 33258 into the microfluidic flow chip, showing that those images were correctly analyzable and blurs were negligible small within 25 μm in depth of microfluidic flow channel for x20 obj. lens. The results indicate the potential of precise identification of cells by simultaneous morphological analysis of bright field/fluorescent images in microfluidic flow.

3Pos224 Multiple-viewpoint analysis of diversity in T cell receptors

Ryo Yokota[1,2], Yuki Kaminaga[3], Tetsuya J. Kobayashi[1,2,3] ([1]*Inst. Ind. Sci., Univ. Tokyo*, [2]*Res. & Edu. Platf. Dyn. Liv. States*, [3]*Sch. Eng., Univ. Tokyo*)

The diversity of T cells in immune system is an important feature to protect our bodies from infection of various pathogens. This diversity is generated by randomization of gene sequences of T cell receptors (TCR) which recognize differences in peptides derived from pathogens. Therefore, investigation of TCR repertoire will probably enable us to know conditions of our health.

In this study, we propose a new method to discriminate the difference in TCR repertoire among different samples. This method is based on not observation frequency of TCR but similarity in amino acid sequences of TCR in itself, and which makes it possible for us to identify the causal sequences for the sample difference.

3Pos225 DNA とナノポアを用いたマイクロメディカルドロップレットシステムの開発

Micro-Medical-Droplet system using DNA and biological nanopore

Moe Hiratani, Masayuki Ohara, Ryuji Kawano (*Tokyo Univ. of Agr. and Tech.*)

MicroRNA (miRNA) has attracted attention as an early diagnostic marker of cancers. To detect miRNA, large-scale apparatuses or time-consuming procedures are required because polymerase chain reaction and gel-electrophoresis are generally used. We have proposed a droplet system with biological nanopores and programmable DNA for the small cell lung cancer (SCLC) therapy, called as Micro-Medical-Droplet (MMD) system. The MMD system conducts three step procedure autonomously: i) miRNA detection, ii) SCLC diagnosis and iii) single-stranded DNA (ssDNA) drug releasing. For the SCLC therapy, ssDNA drugs should be produced 25 times more than miRNA. In this study, we aim at amplification of drug by using enzyme reactions for applying to a drug delivery system.

3Pos226 モータータンパク質で駆動する運動界面の構築と細胞の力学刺激

Dynamic substrate driven by motor proteins for mechanical cellular stimuli

Ryuzo Kawamura, Daiki Uehara, Naritaka Kobayashi, Seiichiro Nakabayashi, Hiroshi Yoshikawa (*Dept. Chem., Saitama Univ.*)

Giving a cell-like property to cell culturing substrates is promising approach for better understanding of the cellular behavior, as the materials surrounding cells are also cells in vivo. Inspired from the dynamic property of the surrounding cells, we propose a new type substrate which can mechanically stimulate culturing cells in a mesoscopic scale from nano to micrometer. With use of microtubule and kinesins, dynamic network of microtubules on a kinesin-coated glass surface was prepared in a cell culture-compatible environment. Seeding cells on this dynamic substrate coupled with ATP hydrolysis, we found that the mechanically stimulated cells showed different morphology compared to those without stimulations.

3Pos227 ポリビニルアルコールゲルで固体化されたバクテリオロドプシンの機能・構造特性に関する分光学的研究

Structural and Functional Characteristics of Bacteriorhodopsin Immobilized with Poly(Vinyl Alcohol) Gel Studied by Spectroscopic Methods

Hikaru Tanaka[1], Yasunori Yokoyama[1], Masashi Sonoyama[2], Koshi Takenaka[1] ([1]*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*, [2]*Div. Mol. Sci., Fac. Sci. Tech., Gunma Univ.*)

We develop a new optical device that applied a photobleaching phenomenon of membrane protein bacteriorhodopsin (bR) by modulating its crystallinity. To immobilize bR with keeping its function is necessary for application to the devices. Poly(vinyl alcohol) gel is good for the matrix in terms of high strength by repetition of freeze-thaw cycle. We have studied structural and functional properties of bR in the gel prepared by the repetition, which is previously unreported. Transient absorption and static CD spectra revealed that the repetition increased its functional efficiency and orientation of purple membrane, i.e. large assembly of bR and lipids, in the gel, whereas functional cycle time was not affected. This shows a strong merit of the gel for application use.

3Pos228 Development of multiple time step integrators in isothermal and isobaric conditions for efficient MD simulations of biological systems

Jaewoon Jung[1,3], Tadashi Ando[2], Yasuhiro Matsunaga[1], Yuji Sugita[1,2,3,4] ([1]*RIKEN AICS*, [2]*RIKEN QBiC*, [3]*RIKEN TMS*, [4]*RIKEN iTHES*)

Molecular dynamics (MD) simulation is a very powerful tool to understand biomolecules phenomena in atomic detail. However, MD requires large number of force calculations due to necessity of using a short time step for simulating biological phenomena occurring on long time scales. Multiple time step integrator has been developed to increase the calculation efficiency without loss of accuracy by using different time steps for “fast” and “slow” forces. Here, we address the two multiple time step integrators: reversible reference system propagator algorithm with Langevin and a stochastic velocity rescaling thermostats/barostats. From the ensemble and dynamical points of view, we will discuss the reliability of the schemes and the maximum time step keeping the accuracy.

3Pos229 フォトクロミック分子を使った紫外光による細胞死の反応機構

Reaction mechanism on cell death due to photochromic molecules upon UV irradiation

Satoshi Yokojima[1,4], Ryuhei Kodama[2], Kimio Sumaru[3], Shinichiro Nakamura[4], Kingo Uchida[2] ([1]*Tokyo Univ. Pharmacy and Life Sci.*, [2]*Ryukoku Univ.*, [3]*AIST*, [4]*Riken*)

A photochromic molecule, diarylenthene with a sulfone group works as the reagent for SO2 gas generation upon UV irradiation. This reaction causes cell death. We here explain the reaction mechanism of the SO2 gas generation upon UV irradiation.

3Pos230 Enhanced efflux activity assists E. coli antibiotic tolerance

Yingying Pu[1], Zhilun Zhao[1], Yingxing Li[1], Jin Zhou[1], Qi Ma[1], Yuehua Ke[1], Yun Zhu[1], Huiyi Chen[2], Hao Ge[1], Yujie Sun[1], Xiaoliang Sunney Xie[1,2], Fan Bai[1] ([1]*Biodynamic Optical Imaging Center, Peking University, Beijing, China*, [2]*Department of Chemistry and Chemical Biology, Harvard University, Cambridge, MA, USA*)

Heterogeneous gene expression in an isogenic bacterial population gives rise to a subgroup, which can tolerate antibiotic attack, termed persisters. Here we use *in vivo* fluorescent imaging to demonstrate that bacterial persisters show less cytoplasmic accumulation of ß-lactam antibiotic, as a result of enhanced efflux activity. Consistently, transcriptome sequencing reveals that TolC exhibits higher expression in persisters. Time-lapse imaging and mutagenesis studies further establish the positive correlation between TolC expression and bacterial persistence. By using a fluorescent membrane potential indicator, persisters are found to undergo more frequent and dramatic membrane potential changes, which may drive the efflux activity enhancement.

Name Index (索引)

名字（Family Name）のアルファベット順にソートしています。すべて、オンラインで入力されたデータのまま、表示しています。

Name Index

第53回 日本生物物理学会年会

浜松ホトニクス株式会社 ランチョンセミナー

◇ プログラムNo.1LG

◇ 日時:2015年9月13日(日) 11:50 ～ 12:40

◇ 会場:G会場 (レクチャーホール)

演題1

蛍光・化学発光イメージングに相応しいカメラの選び方

永井 健治 先生
(大阪大学 産業科学研究所 生体分子機能科学研究分野)

【 要旨 】

蛍光タンパク質や化学発光タンパク質をエンジニアリングすることで、細胞や組織、個体内で繰り広げられている様々な生命現象を可視化することが可能なプローブが開発されてきた。これら光プローブのシグナルを検出するには当然のことながらカメラが無くてはならない。今最も汎用されている科学計測用カメラとしてsCMOSとEM-CCDが挙げられるが、それぞれどのような利点･欠点があるのかを把握して使用しないと、捉えられる現象も捉えられず、ただの宝の持ち腐れになってしまう。本セミナーでは我々の研究室で開発した超解像イメージング用光スイッチング蛍光プローブと膜電位イメージング用高光度化学発光プローブを取り上げ、これらのシグナルを可視化するのに要求されるスペックを解説しながら、相応しいカメラの選び方について高速、広視野、高画素数、多波長をキーワードに考察する。

演題2

業界をリードする浜松ホトニクスの最新CMOSカメラ技術

戸田 英児
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第５３回日本生物物理学会年会

株式会社菱化システム ランチョンセミナー

バイオインフォマティクス技術の開発と応用

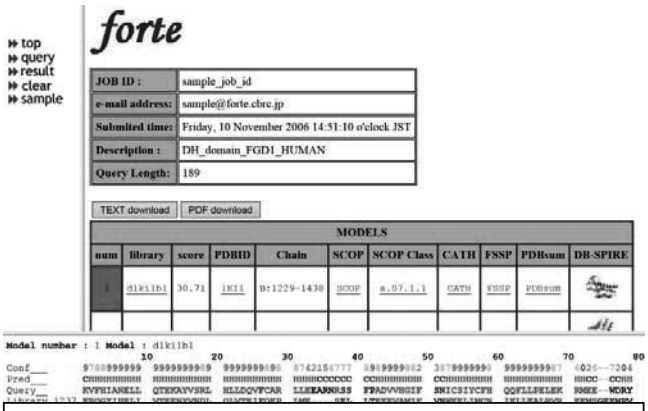
日時：９月１３日（日）　11:50 ～ 12:40

場所：Ｈ会場（大講義室 A）

構造バイオインフォマティクス技術を利用したタンパク質の機能解析

国立研究開発法人産業技術総合研究所　創薬基盤研究部門　**富井 健太郎**

タンパク質立体構造情報とその解析あるいは予測手法は、機能未知タンパク質のドメイン推定や類縁関係の類推、基質結合部位の予測等を通して、生物学的研究の効率化や推進に活かされています。われわれの研究グループでは、創薬等の支援に向け、タンパク質立体構造予測、相互作用部位予測等の構造バイオインフォマティクス技術の開発及び高度化とそれ等を利用した生物学的研究の支援を行っています。本発表では、アミノ酸配列の情報解析に基づくタンパク質立体構造予測法をはじめとする開発技術の効果と、それ等技術の創薬標的候補タンパク質の解析/探索等への応用例について紹介します。



タンパク質類似性検索ツール forte の計算画面
http://forteprtl.cbrc.jp/forte/

Structure-Based Drug Design (SBDD) におけるデータ管理

株式会社菱化システム 科学技術システム事業部 木村 嘉朗

SBDD の研究で利用される多様かつ大量のデータを管理するには、多くの手間と時間が必要です。統合計算化学システム MOE の SBDD データ管理ツール MOE Project では、タンパク質立体構造とその構造アノテーションや、リガンド構造とそのプロパティ・データを自動的に収集して、一つのデータベース・ファイルにまとめます。本セミナーでは、この MOE Project を用いた SBDD のデータ管理機能を紹介します。

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PDBj
Protein Data Bank Japan

Protein Data Bank Japan

Platform for Drug Discovery, Informatics, and Structural Life Science

Luncheon Seminar at the 53rd Annual Meeting of Biophysical Society of Japan

September 13 (Sun), 11:50-12:40, Room I（Lecture Room B）

Rapid advancement of high-resolution cryo-EM intrudes a database of atomic coordinates.

急速な高分解能クライオ電子顕微鏡の進歩による原子座標データベースの増加

Kenji Iwasaki, Institute for Protein Research, Osaka University

Almost every week the near-atomic resolution structures of various proteins determined by single-particle reconstruction (SPR) cryo-EM are reported in high profile journals, corresponding with a dramatic increase in the deposition of atomic coordinates into the PDB or EMDB. This revolution has been caused by the development of direct electron detection cameras. The rapid development of cryo-EM will be introduced in this seminar, along with a method developed in our laboratory: the hybrid approach, which combines EM, computer simulation, and crystal structures.

New deposition system and a validation tool of Protein Data Bank

Protein Data Bank の新しい登録システムと構造評価ツール

Atsushi Nakagawa, Institute for Protein Research, Osaka University

Because of recent advances of structure determination techniques, such as X-ray crystallography, NMR, cryo-EM etc, number of protein structures determined is dramatically increased. Furthermore, huge macromolecular complexes have been solved at atomic resolution. In the middle of 70s, when Protein Data Bank started to archive the atomic coordinates of proteins, only several tens of structures were deposited. The legacy PDB format has limited capabilities, since it is based on the techniques of computer and X-ray crystallography at that time. For example, number of chains is defined as one-byte capital character and number of residues is limited to 999. Large biological macromolecular assemblies, for example ribosome, exceeded these limitations. PDB decided to extend the PDB format to a new format, named PDBx/mmCIF, with much more capabilities. The PDBx/mmCIF is an extension of the legacy format, but it is more flexible. In addition to the new format, the new deposition system, which includes a validation tool, is now working in all PDB deposition sites since last January. All X-ray crystal structure must be deposited via the new deposition system and quality of the data can be assured with this new deposition system.

Introduction to Structural Life Science Data Cloud

構造生命科学データクラウドの紹介

Akira R. Kinjo, Institute for Protein Research, Osaka University

While protein structures provide detailed information about their molecular functions, it is often necessary to integrate such information in a broader context in order to understand the biological functions. In the Platform for Drug Discovery, Informatics, and Structural Life Science, we have been developing the VaPRoS system that amalgamates a wide range of information such as genomes, gene expression, protein structures, protein complexes, and more. In this seminar, I will introduce some of the basic functionalities of the VaPRoS system for the end user.

Protein Data Bank Japan
http://pdbj.org/

Institute for Protein Research, Osaka University,
3-2 Yamadaoka, Suita, Osaka 565-0871, Japan
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第53回日本生物物理学会年会ランチョンセミナー

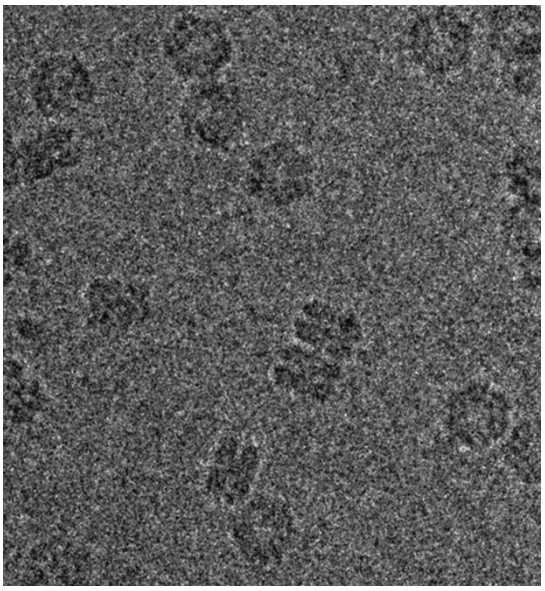
The latest developments and improvements in Cryo-TEM workflow solutions

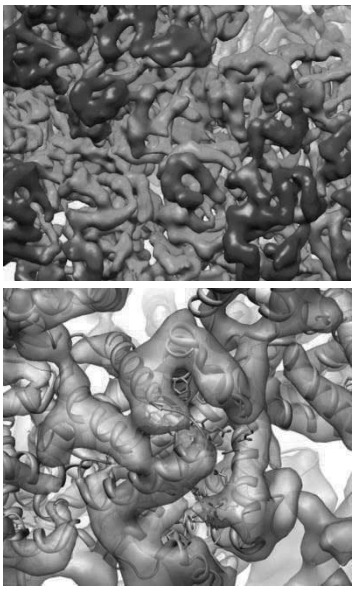
日時: 9月14日(月) 11:30 – 12:20

場所: A会場 （金沢大学　自然科学本館　大会議室）

演者: 葦原　雅道 （日本エフイー・アイ株式会社）

生体高分子立体構造解析におけるクライオ電子顕微鏡法は近年、急速な進展を遂げた。 単離精製した生体高分子をターゲットとする単粒子解析法では、電子顕微鏡の密度マップのみから原子モデル構築が可能な分解能での構造解析がルーティン化しつつある。
このようなクライオ電子顕微鏡法の急速な進展の背景には技術的なブレークスルーがある。電子顕微鏡の自動化、高感度、高S/Nの画像記録を実現するダイレクトディテクター、位相板などがあげられる。
クライオ電子顕微鏡法の高分解能化により、生体高分子の立体構造解析手法としてX線結晶構造解析法やNMR法と並ぶ代表的な手法となった。本セミナーでは、クライオ電子顕微鏡法の実験の流れから技術的革新まで様々な構造解析例とともに紹介する。





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オリンパス(株)ランチョンセミナー

細胞膜の幾何学と物理学にもとづく細胞運動の理解

日　時

9月14日［月］　※昼食時間帯にて実施予定。

会　場

金沢大学 角間キャンパス 自然科学本館

演　者

伊藤 俊樹 先生

神戸大学バイオシグナル研究センター 生体膜機能研究分野 教授

伊藤 俊樹（いとう としき）先生 略歴

1994年　東京大学理学部生物化学科 卒業
1999年　東京大学大学院理学系研究科生物化学専攻博士課程 修了
2000年　東京大学医科学研究所 助手
2003年　Yale大学医学部 博士研究員
2005年　東京大学医科学研究所 助教授
2007年　神戸大学大学院医学研究科 准教授
2013年　神戸大学バイオシグナル研究センター　教授

- 主要論文 -　*corresponding author

Tsujita, K., Takenawa, T., Itoh, T.*
Feedback regulation between plasma membrane tension and membrane-bending proteins organizes cell polarity during leading edge formation.
Nat. Cell Biol. 17 (6), 749-758 (2015)

Hasegawa, J., Tokuda, E., Tenno, T., Tsujita, K., Sawai, H., Hiroaki, H., Takenawa, T., Itoh, T.*
SH3YL1 regulates dorsal ruffle formation by a novel phosphoinositide-binding domain.
J. Cell Biol. 193 (5), 901-916 (2011)

Itoh, T.*, Hasegawa, J., Tsujita, K., Kanaho, Y. Takenawa, T.*
The tyrosine kinase Fer is a downstream target of the PLD-PA pathway that regulates cell migration.
Science Signaling 2 (87), ra52 (2009)

Itoh, T., Erdmann, K. S., Roux, A., Habermann, B., Werner, H. De Camilli, P.
Dynamin and the Actin Cytoskeleton Cooperatively Regulate Plasma Membrane Invagination by BAR and F-BAR Proteins.
Dev. Cell 9 (6), 791-804 (2005)

Itoh, T., Koshiba, S., Kigawa, T., Kikuchi, A., Yokoyama, S. Takenawa, T.
Role of the ENTH domain in phosphatidylinositol-4,5-bisphosphate binding and endocytosis.
Science 291 (5506), 1047-1051 (2001)

全反射顕微鏡や光ピンセットなどのイメージング手法を実験に取り入れ、細胞膜の物理的シグナルによる細胞運動のメカニズム解明に取り組んでいる神戸大学 伊藤 俊樹教授。本ランチョンセミナーでは伊藤 俊樹教授を講師にお迎えし、今年５月に「Nature Cell Biology」で発表された研究成果などを中心に、セルバイオロジーにおける最新のイメージング技術についてご紹介します。

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第 53 回生物物理学会年会　ランチョンセミナー
オックスフォード・インストゥルメンツ株式会社

日時
9 月 14 日(月)　11:30～12:20

会場
レクチャーホール(G 会場)

演者
菅澤 祐昭　**(オックスフォード・インストゥルメンツ(株)アサイラム リサーチ事業部)**
渡邉 朋信 先生　**(国立研究開発法人理化学研究所生命システム研究センター 先端バイオイメージング研究チーム　チームリーダー)**

要旨

AFM(原子間力顕微鏡)は、”生きたままの状態“で観察できるだけでなく、力学物性も計測することも特長の一つである。アサイラム リサーチの AFM は、先進的な液中観察技術と、定量性を追求したフォース測定技術で、業界をリードしている。今回は、これらの技術の詳細と応用例について紹介する。

高感度 EMCCD カメラ iXon の出現は、それまでの一分子追跡技術を大きく進歩させた。高感度と高フレームレートが共に達成されたからである。講演者は、この利点を利用し、細胞内でのモータータンパク質のナノ運動の追跡、さらには、マウス個体内での追跡にも成功してきた。本ランチョンセミナーでは、講演者の iXon と共にあった技術開発について紹介する。

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第53回日本生物物理学会年会

ニコンインステック　ランチョンセミナー

超解像顕微鏡を用いた Xist RNA のイメージング

増井 修 先生

理化学研究所　統合生命医科学研究センター
免疫器官形成研究グループ

日 時　2015年 9月15日（火）12:15～13:05

会 場　金沢大学角間キャンパス自然科学本館
H会場（大講義室A）

最近の超解像蛍光顕微鏡の実用化により、対象物の構造をこれまでよりも詳細に解析することが可能となった。超解像技術には様々な原理に基づいた物が存在しているが、それらの中でも構造化照明法（SIM）は、顕微鏡にそれほど詳しくない生物学研究者にも比較的扱いやすいシステムであると言える。一方、RNA 分子を蛍光シグナルとしてイメージングするためには、RNA FISH 法のように変性剤を使用した浸潤的な方法がこれまでに用いられてきたが、細胞内構造への変性剤の影響を排除できないという短所があった。我々は最近、RNA に MS2 や Bgl などのステムループを挿入してタグ化することで、非変性環境下での免疫染色や、生細胞でのライブイメージングによる解析を実現した。今回のセミナーではこれらの非浸潤的な RNA 可視化法を用いて Xist RNA を SIM により超解像解析した例について報告したい。

****

第53回 日本生物物理学会年会

オプトライン ランチョンセミナー

光スイッチング蛍光タンパク質が実現する次世代超解像イメージング

大阪大学　産業科学研究所　生体分子機能科学研究分野

和沢 鉄一 先生、　**新井 由之** 先生、　**永井 健治** 先生

日　時　9月15日（火）　12: 15 ～ 13 : 05

会　場　F 会場（金沢大学角間ｷｬﾝﾊﾟｽ自然科学本館内）

光の回折限界を超える空間分解能での観察を可能にする超解像顕微法により、従来の光学顕微鏡では見えなかった細胞内の複雑な微細構造を非侵襲で可視化できるようになってきた。このような超解像顕微法として、1分子イメージングの拡張であるPALMや、試料の照明パターンを制御する方法であるRESOLFTなどが開発されている。しかしながら、従来の超解像顕微法は、生細胞イメージングに適した蛍光プローブがない、撮像時のフレームレートがリアルタイム観察に及ばない、強力な光の照射によって細胞に対して著しい光毒性が発生する、顕微鏡システムが高価になる、といった問題があった。このため、生命科学研究において超解像イメージングは未だ広く普及していないのが現状である。我々は、簡素な光学系による生細胞に優しいリアルタイム超解像イメージングの実現を目指して、新規光スイッチング蛍光タンパク質の開発と、その特性を如何なく利用した超解像顕微法の開発に取り組んできた。

本セミナーでは、偏光照明による“細胞に優しい”超解像顕微法、並びに多波長Compactレーザー光源を用いた“超簡単”超解像顕微法について紹介する。

RIBM

第 53 回日本生物物理学会年会 ランチョンセミナー

2015 年 9 月 15 日（火）12：15 – 13：05

会場：G 会場（レクチャーホール）

演者 1　高速 AFM　～どう使うか、何が分かるか～

金沢大学 理工研究域 数物科学系 教授

安藤敏夫 先生

高速 AFM は低侵性に優れ、タンパク質分子が機能している時の動的な姿・形をサブ分子分解能、サブ 100ms の時間分解能で、直接観察できる唯一の顕微鏡である。蛍光顕微鏡と違い、染色は不要である。また、生細胞で起こる動的な現象も動画観察可能である。

この講演では、高速 AFM の原理、特徴、使い方を分かりやすく説明し、得られた動画映像とそこから得られた発見などを紹介する。

演者 2　高速原子間力顕微鏡　PS-NEX のご紹介

－プローブスキャン型動画 AFM ×　蛍光顕微鏡によるナノライブイメージング－

株式会社 生体分子計測研究所

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本学会の連絡先は下記の通りです.

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   E-mail bpsjp@biophys.jp
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   岩波書店一ツ橋別館 4F　株式会社エー・イー企画
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   http://www.biophys.jp

本誌記事の動物実験における実験動物の扱いは,
所属機関のルールに従っています.

生物物理 SEIBUTSU BUTSURI
日本生物物理学会 THE BIOPHYSICAL SOCIETY OF JAPAN

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