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生物物理

SEIBUTSU BUTSURI

SUPPLEMENT 1-2
Vol.59

第57回年会予稿集

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　https://www.biophys.jp

第 57 回日本生物物理学会年会

The 57th Annual Meeting of the Biophysical Society of Japan

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開催概要／General Information

The 57th Annual Meeting of the Biophysical Society of Japan (BSJ2019)
第 57 回生物物理学会年会（2019 年度）

会期／Period

2019 年 9 月 24 日（火）－26 日（木）
September 24 (Tue.) – 26 (Thu.), 2019

会場／Venue

シーガイアコンベンションセンター
（〒 880-8545 宮崎市山崎町浜山）
Seagaia Convention Center (Hamayama, Yamazaki-cho, Miyazaki, 880-8545, Japan)

年会長／Chair

永井 健治（大阪大学産業科学研究所）
Takeharu Nagai (The Institute of Scientific and Industrial Research, Osaka University)

Website http://www2.aeplan.co.jp/bsj2019

※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）

※下記サイトからダウンロードいただけます。
http://www.biophys.jp/dl/pro/57th_proceedings.pdf
ID: ambsj57　Password: miyazaki2019
※スマートフォン・タブレット端末向けのプログラム検索・要旨閲覧アプリは，2019 年 9 月 18 日（水）公開予定です。

開催にあたって

第 57 回年会　年会長
永井 健治
（大阪大学 産業科学研究所 生体分子機能科学研究分野）

さて！第 57 回日本生物物理学会年会は 2019 年 9 月 24 日（火）から 26 日（木）の 3 日間, 宮崎市のフェニックス・シーガイア・リゾートにて開催する運びとなりました。

何故宮崎で開催なのか？

それはひとえに**「年会参加者の，年会参加者による，年会参加者のための交流の場」を実現したいから**に他なりません。そのために本年会のコンセプトとして「祭り」を掲げました。ご存知のように，生物物理学会年会では，一年に一度，会員間の研究情報の交換を目的に，全国の大学および国・公立・民間の研究機関等より約 1,500 人の研究者が一堂に会し発表と討論が行われております。1,500 人を一堂に集めるには, それなりのスペースが必要です。

今回，参加者 1,500 人全員で「祭り」を行うべく，**1 フロアで全員収容可能な会場を用意**しました。それが 2000 年に G8 サミットが開催された宮崎フェニックス・シーガイア・リゾートです。都会の雑踏から離れ，まさに，G8 サミットのごとく，他のことにうつつを抜かすことなく，3 日間，思う存分サイエンスの祭りに専念できるのです。サイエンスに専念？会場周辺に何もあらへんやん。昼ごはんはどないすんねん？

ご安心ください！会場外に昼食の場を探す必要も無いように，**参加者全員がお昼ご飯にありつけるだけのランチョンセミナーを開催**します。特に本年会では，ランチョンセミナーをバイオフィジクスセミナーと改名し，より科学的な企業紹介をみなさまに提供致します。

1 フロアに 1,500 人？企業展示ブースは全く別の部屋に設置されるんか？行き来が大変やん？
ご安心ください！**企業展示ブースはフロアのど真ん中**にあり，ポスター会場に取り囲まれています。ポスターセッションで科学的議論に疲れたら，気軽に企業展示ブースに足を運ぶことが可能です。

シーガイアは宮崎市街地からめっちゃ離れとるやん。会場と市内ホテルとの往復がめっちゃしんどいんちゃう？

ご安心ください！**シェラトン・グランデ・オーシャンリゾートホテルに極めてリーズナブルな価格で宿泊**できます。是非ご利用ください！

いや，そやかて年会参加費が高すぎて参加でけるかどうか分からへんわ。

ご安心ください！**トラベルグラント**を設けました。特に学生や若手研究者の皆様，是非ご応募ください！みなさまの不安は全て取り除きました。まさに至れり尽くせりの年会です。

なんと！それだけではありません！本年会の懇親会は会場に隣接する広大な野外芝生広場で開催いたします。まさに**年会トップページのイラストを地で行く「お祭り」懇親会**です。この懇親会では新しい試みとして，研究者と企業の方との交流を促進する**情報交換特別展示場**を設けます。きっとご自身の研究に有用な情報が今まで以上に得られることでしょう。その他にも懇親会では「あっ！」と言わせる仕掛けが目白押しです。

極めつけはなんといっても，シーガイア周辺のリゾートアクティビティです。年会後に，ゆっくりと宮崎のさわかやかな風を感じながらフェニックスカントリークラブでゴルフを楽しむもよし。宮崎神宮で論文アクセプト祈願をするのもよし。日南海岸でサーフィン三昧もよし。ご自身の色々と相談の上でお楽しみいただければと思います。

もちろん, **研究発表そのものにもぬかりはありません**。肝心の昼間のセッションではシンポジウムやポスターセッションに加え，生物物理学の基礎から最新情報まで全体を俯瞰できるカレントトピックセッション，企業トーク，キャリアセミナーなどみなさんの知的好奇心をくすぐる企画が満載です。

初秋の宮崎は，夏のギラギラした暑さが和らぎ，さわやかな季節です。もう少しだけ season in the sun! 本年会は真夏さながらの熱気できっと次のサイエンスにつながる相転移を起こすこと間違いなし。**早朝から深夜，そしてアフター学会までとことん**生物物理学の進歩に触れ，新たな切り口から生命現象を討議しませんか？

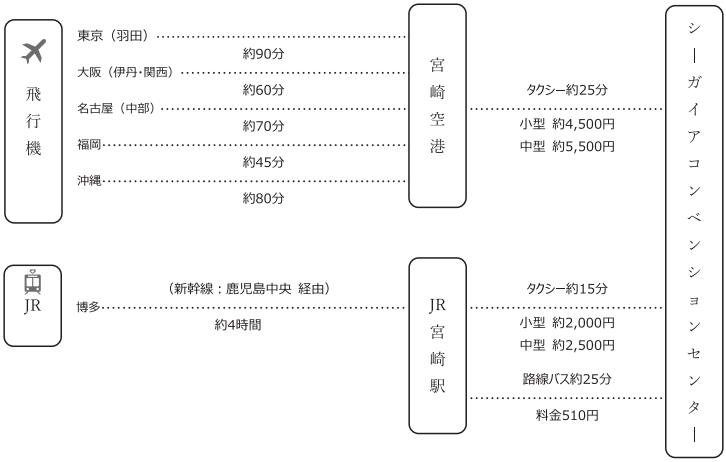
参加者の皆様が存分に楽しんで頂ける魅力満載の年会になるよう年会実行委員会一同で準備を進めています。多数の演題応募とご参加を心よりお待ちしております。

祭りだ！祭りだ！サイエンスのお祭りだ！
みんな宮崎に集まれ！

第57回日本生物物理学会年会実行委員会
Organizing Committee

年会長		Chair	
永井 健治	（大阪大学）	Takeharu Nagai	(Osaka University)
実行委員長		**Director General**	
宮田 真人	（大阪市立大学）	Makoto Miyata	(Osaka City University)
副実行委員長		**Director of Accounting**	
今田 勝巳	（大阪大学）	Katsumi Imada	(Osaka University)
プログラム委員長		**Director of Program**	
城口 克之	（理化学研究所）	Katsuyuki Shiroguchi	(RIKEN)
実行委員		**Members**	
池谷 裕二	（東京大学 / 大阪大学）	Yuji Ikegaya	(University of Tokyo / Osaka University)
伊東 祐二	（鹿児島大学）	Yuji Ito	(Kagoshima University)
上田 昌宏	（大阪大学）	Masahiro Ueda	(Osaka University)
内山 進	（大阪大学）	Susumu Uchiyama	(Osaka University)
大上 雅史	（東京工業大学）	Masahito Ohue	(Tokyo Institute of Technology)
岡田 眞里子	（大阪大学）	Mariko Okada	(Osaka University)
岡田 康志	（東京大学／理化学研究所）	Yasushi Okada	(RIKEN / University of Tokyo)
樺山 一哉	（大阪大学）	Kazuya Kabayama	(Osaka University)
神田 元紀	（理化学研究所）	Genki Kanda	(RIKEN)
菊地 和也	（大阪大学）	Kazuya Kikuchi	(Osaka University)
昆 隆英	（大阪大学）	Takahide Kon	(Osaka University)
榊原 陽一	（宮崎大学）	Yoichi Sakakibara	(University of Miyazaki)
白上 努	（宮崎大学）	Tsutomu Shiragami	(University of Miyazaki)
水光 正仁	（宮崎大学）	Masahito Suiko	(University of Miyazaki)
谷 知己	（産業技術総合研究所）	Tomomi Tani	(AIST)
中瀬 生彦	（大阪府立大学）	Ikuhiko Nakase	(Osaka Prefecture University)
原田 慶恵	（大阪大学）	Yoshie Harada	(Osaka University)
藤井 律子	（大阪市立大学）	Ritsuko Fujii	(Osaka City University)
藤田 克昌	（大阪大学）	Katsumasa Fujita	(Osaka University)
細川 千絵	（大阪市立大学）	Chie Hosokawa	(Osaka City University)
松浦 友亮	（大阪大学）	Tomoaki Matsuura	(Osaka University)
南野 徹	（大阪大学）	Tohru Minamino	(Osaka University)
吉村 成弘	（京都大学）	Shige H Yoshimura	(Kyoto University)
和田 啓	（宮崎大学）	Kei Wada	(University of Miyazaki)
渡邊 朋信	（理化学研究所）	Tomonobu M Watanabe	(RIKEN)

会場へのアクセス

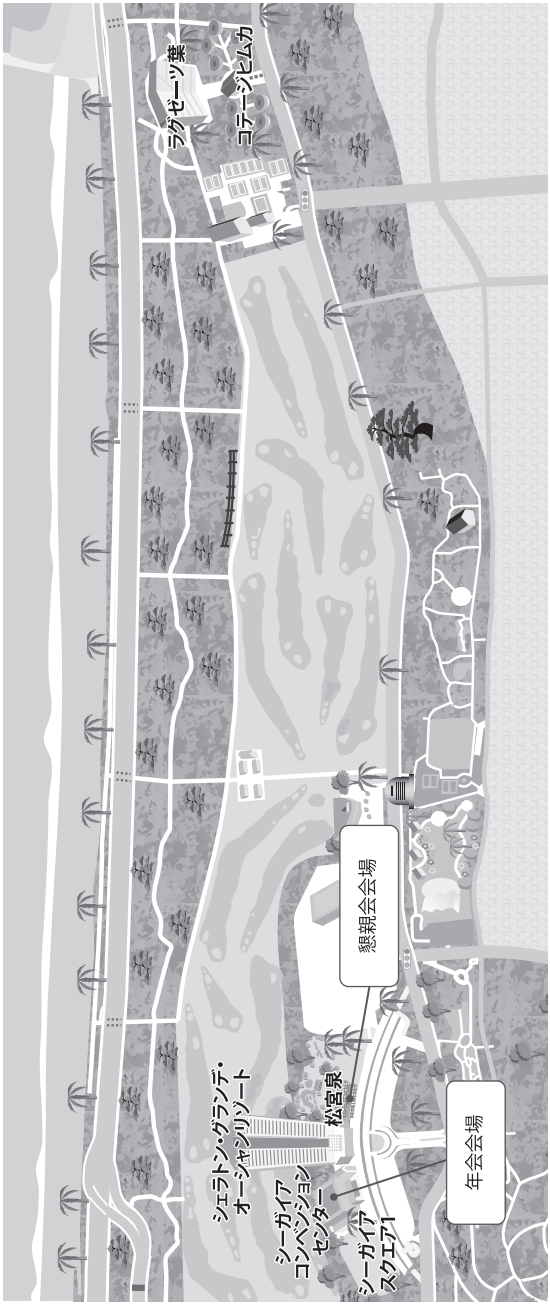


※宮崎空港から会場までは乗合タクシーをご用意しております。
　空港の年会専用の乗合タクシー受付にお越しください。

※年会会期中は，貸切バスを運行する予定です。
　・朝　宮崎駅→会場
　・年会終了後　会場→宮崎駅

※年会最終日，クロージングリマーク終了後，空港行の貸切バスを運行します。

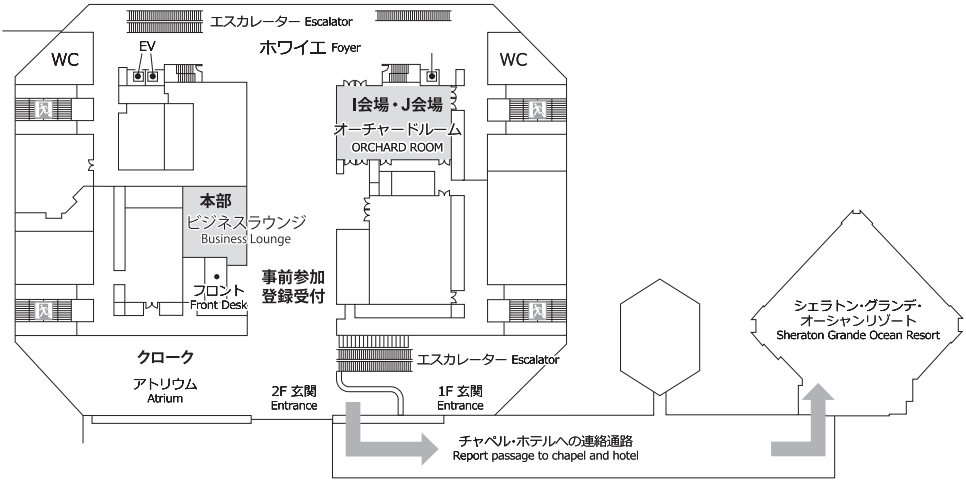
フェニックス・シーガイア・リゾート エリアマップ



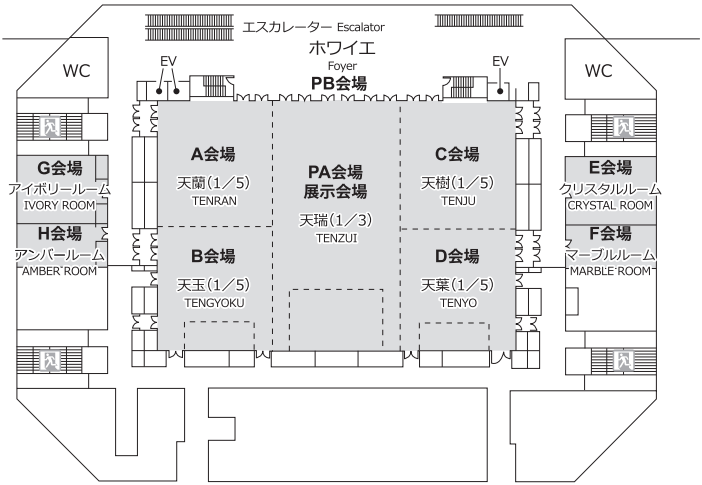
※ラグゼーツ葉・コテージヒムカから年会会場のあるコンベンションセンターまでは約3Kmありますので，園内シャトルバスをご利用ください。

会場図

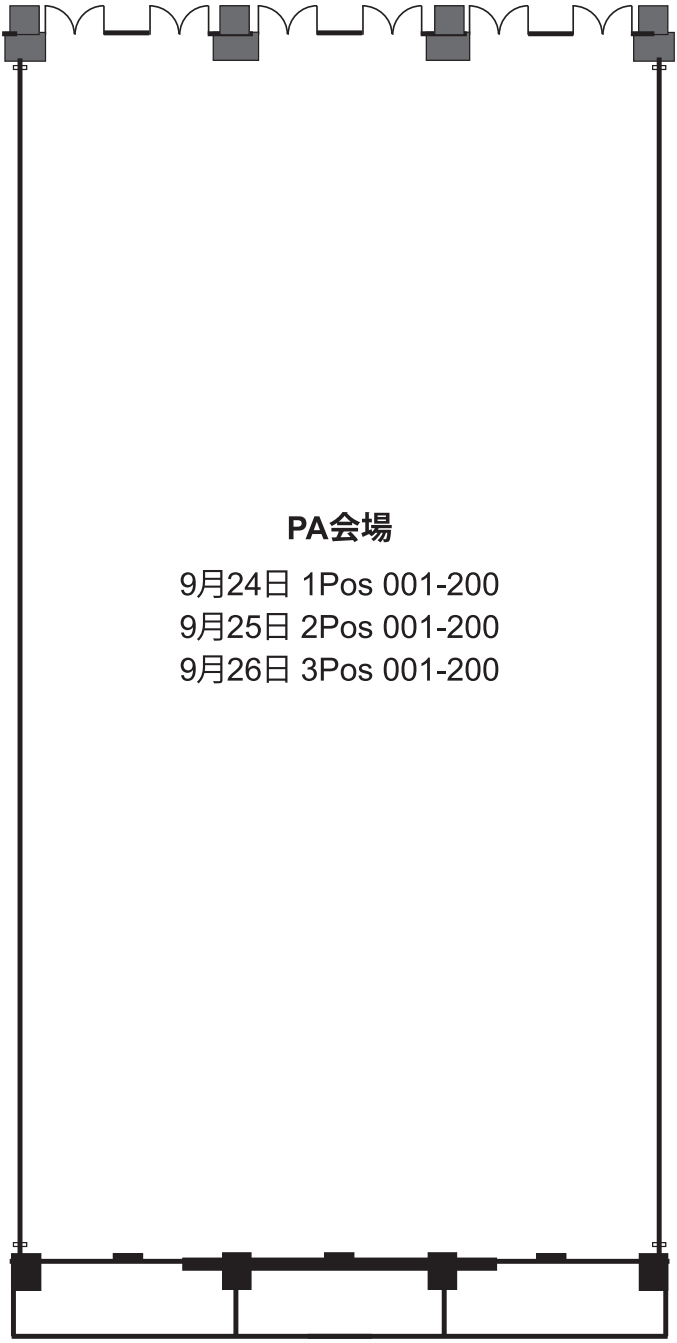
2nd Floor



4th Floor

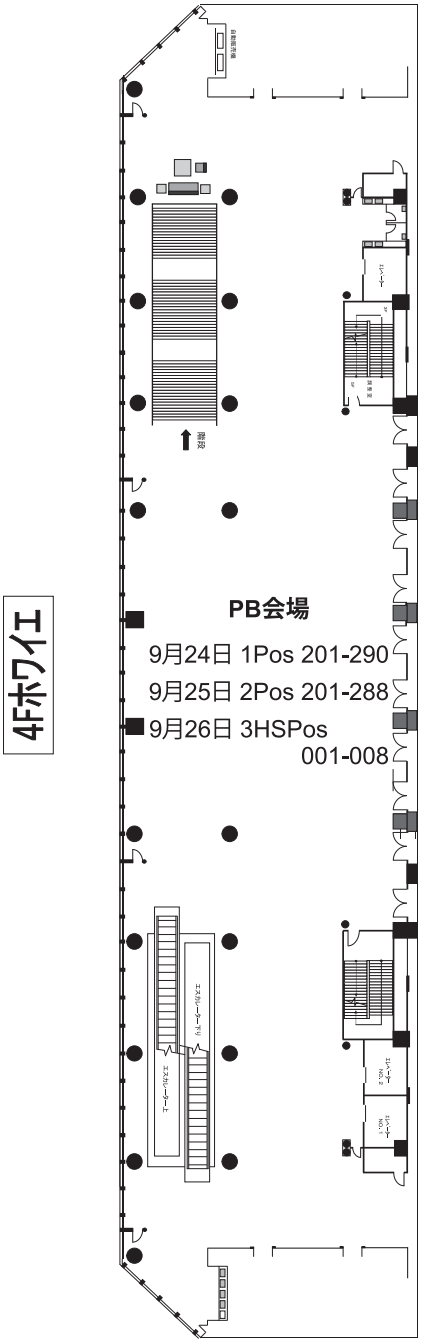


ポスター会場



PA会場

9月24日 1Pos 001-200
9月25日 2Pos 001-200
9月26日 3Pos 001-200


4Fホワイエ
PB会場
9月24日 1Pos 201-290
9月25日 2Pos 201-288
9月26日 3HSPos
001-008
エスカレーター
エスカレーター
階段

■2019年9月23日（月・祝）：年会前日

階	部屋名	会場名	8	9	10	11	12	13
2F	オーチャード南	J会場						

■2019年9月24日（火）：年会1日目

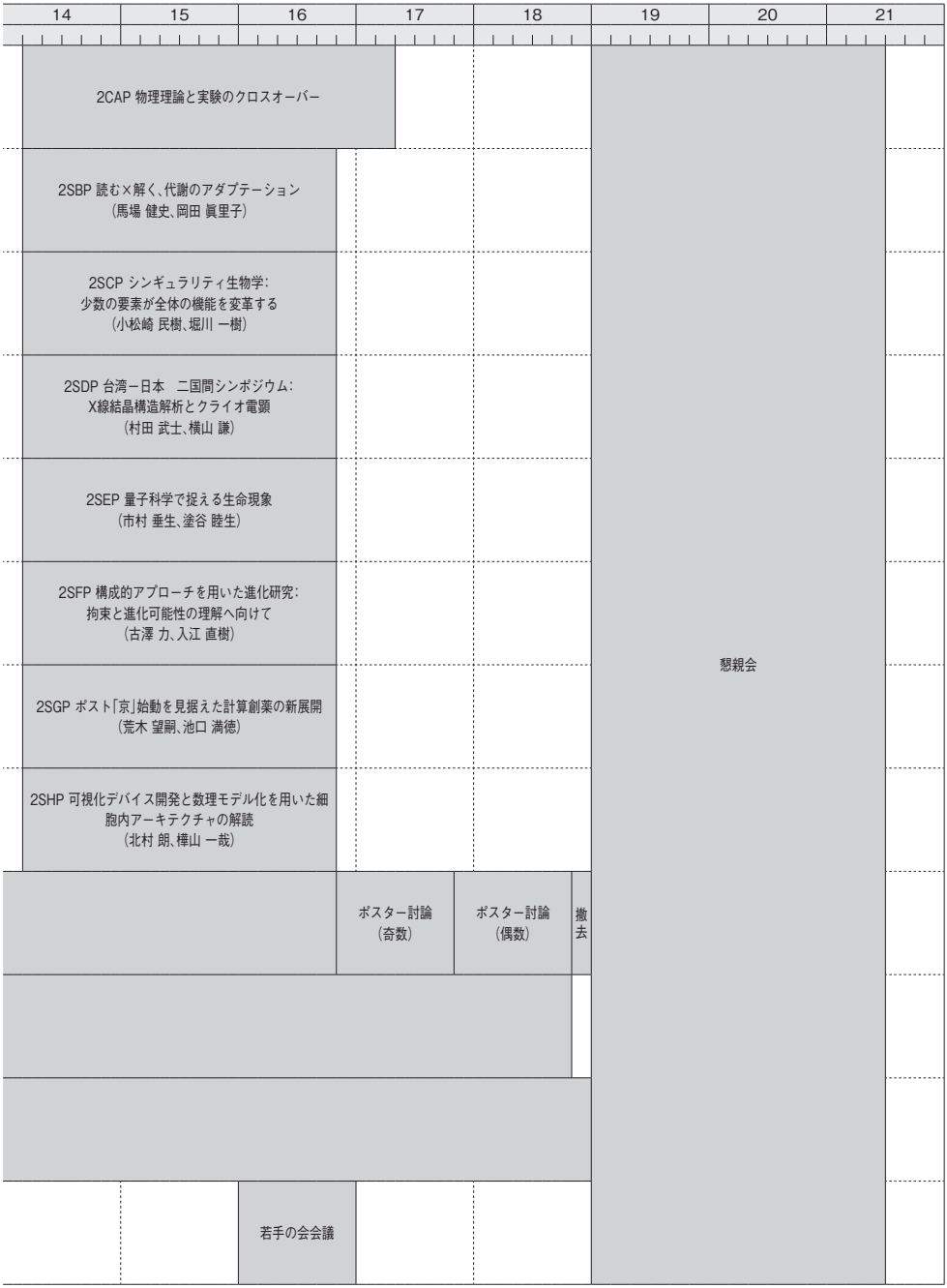
階	部屋名	会場名	8	9	10	11	12	13
4F	天蘭	A会場		1YA 若手招待講演		ソーラボジャパン株式会社	論文賞講演会	
	天玉	B会場	ヨダカ	1SBA ゲノム機能発現の統合的理解に向けた多角的アプローチ（森 貴治、関根 俊一）		株式会社ニコンインステック		
	天樹	C会場	ナニオン	1SCA 階層を超えた柔軟な場と空間の活用：生命システムが持つ可能性を探る（佐藤 佑介、森田 雅宗、鈴木 勇輝）		株式会社オプトライン		
	天葉	D会場	スペクトリス	1SDA クライオ電子顕微鏡でできること、できないこと ー構造生命科学の最先端ー（中川 敦史、吉川 雅英）		浜松ホトニクス株式会社		
	クリスタルルーム	E会場		1SEA 遺伝子制御の原理に迫るクロマチン動態の物理学（伊藤 由馬、木村 暁）		日本ウォーターズ株式会社		
	マーブルルーム	F会場		1SFA 生体機能の「ありのまま」の可視化と理解へ ～共鳴する生命現象と光技術～（宮脇 敦史、根本 知己）		ヨダカ技研株式会社		
	アイボリールーム	G会場	ソーラボ	1SGA 筋・血管系のマルチスケールメカノバイオロジーの最前線（岩城 光宏、原　雄二）		日本蛋白質構造データバンク（PDBj）		
	アンバールーム	H会場	INTEGRA	1SHA 静水圧刺激により生命機能を操作する（畑 宏明、西山 雅祥）		DKSHジャパン株式会社		
	天瑞・ホワイエ	PA会場/PB会場	貼付	ポスター掲示				
	天瑞	展示会場			機器・試薬・書籍展示			
2F	オーチャード北	I会場				キャリア支援説明会		
	オーチャード南	J会場			BPPB誌編集委員会	出版委員会	2019年度第2回理事会	

14	15	16	17	18	19	20	21
ウェブサイト編集委員会			生物物理誌編集委員会				

14	15	16	17	18	19	20	21
1CAP タンパク質の構造・機能・デザイン							
1SBP オーストラリア－日本 交流シンポジウム: 生体分子相互作用と介在する力にフォーカスした生物物理学の挑戦 (西坂 崇之、Marc Kvansakul)							
1SCP 生物物理で見る脳神経回路 (冨永 貴志、Bernd Kuhn)							
1SDP 蛋白質の溶液物性計測の現状と課題 (内山 進、谷中 冴子)							
1SEP さきがけ「1細胞」は何をやっている？ 1細胞研究の醍醐味と技術革新 (城口 克之、鈴木 団)							
1SFP 高感度水素検出による 生体内化学反応の制御を目指して (田中 伊知朗、石北 央)							
1SGP 高次元データ駆動科学と計測インフォマティクスによる分子観察の新展開 (木川 隆則、松永 康佑)							
1SHP GPCRによる多様な情報伝達機構を解き明かす構造－機能相関研究の新展開 (片山 耕大、寿野 良二)							
		ポスター討論 (奇数)	ポスター討論 (偶数)	撤去			
		キャリア支援説明会　個別相談会					
		若手奨励賞 選考委員会					

■2019年9月25日（水）：年会2日目

階	部屋名	会場名	8	9	10	11	12	13
4F	天蘭	A会場	東京インスツルメント	2CAA 細胞の生物物理学		分野別専門委員会	会員総会・総会シンポジウム	
	天玉	B会場		2SBA 分子夾雑のススメ（田端 和仁、三好 大輔）		オリンパス株式会社		
	天樹	C会場		2SCA 分子構造ビッグデータの時代のバイオインフォマティクスの挑戦（白井 剛、寺田 透）		マルバーン・パナリティカル事業部（スペクトリス株式会社）		
	天葉	D会場		2SDA 生体分子機械の非平衡エナジェティクス（鳥谷部 祥一、Chun-Biu Li）		サーモフィッシャーサイエンティフィック株式会社		
	クリスタルルーム	E会場		2SEA 放射光利用生物物理研究の最前線（岩本 裕之、関口 博史）		シグマ光機株式会社		
	マーブルルーム	F会場		2SFA 光操作による生命機能解析（七田 芳則、塚本 寿夫）		株式会社アントンパール・ジャパン		
	アイボリールーム	G会場		2SGA 電子・熱・化学エネルギーの生体内伝達と地域社会実装に向けた基盤研究（和田 啓、榊原 陽一）		ベックマン・コールター株式会社		
	アンバールーム	H会場	カンタム	2SHA タンパク質のダイナミックレスポンスに関わる未解決問題への挑戦（鷹野 優、米澤 康滋）		SCIEX		
	天瑞・ホワイエ	PA会場/PB会場	貼付	ポスター掲示				
	天瑞	展示会場			機器・試薬・書籍展示			
2F	オーチャード北	I会場			キャリア支援説明会			
	オーチャード南	J会場						


14
15
16
17
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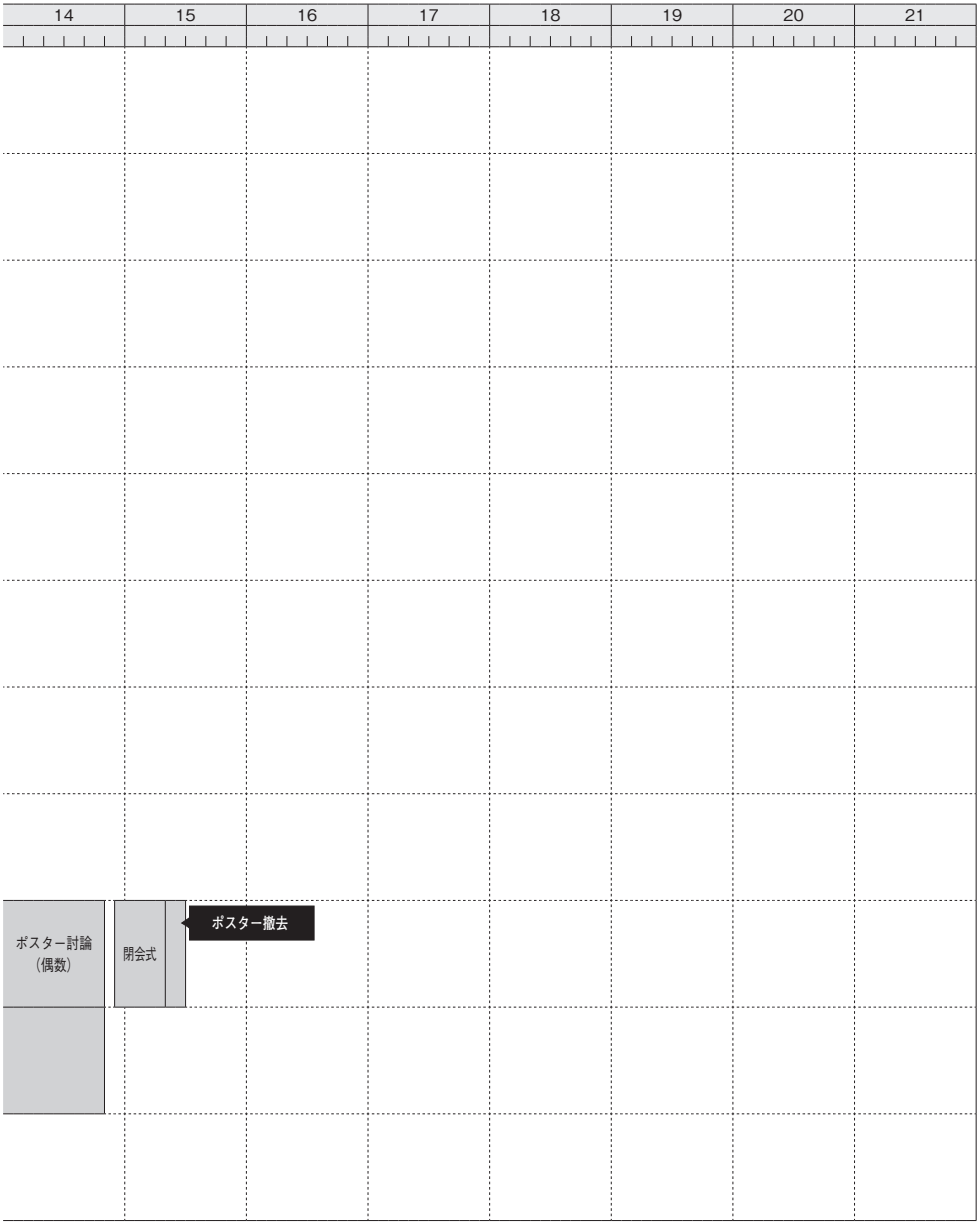
19
20
21
2CAP 物理理論と実験のクロスオーバー
2SBP 読む×解く、代謝のアダプテーション
（馬場 健史、岡田 眞里子）
2SCP シンギュラリティ生物学:
少数の要素が全体の機能を変革する
（小松崎 民樹、堀川 一樹）
2SDP 台湾－日本　二国間シンポジウム:
X線結晶構造解析とクライオ電顕
（村田 武士、横山 謙）
2SEP 量子科学で捉える生命現象
（市村 垂生、塗谷 睦生）
2SFP 構成的アプローチを用いた進化研究:
拘束と進化可能性の理解へ向けて
（古澤 力、入江 直樹）
2SGP ポスト「京」始動を見据えた計算創薬の新展開
（荒木 望嗣、池口 満徳）
2SHP 可視化デバイス開発と数理モデル化を用いた細
胞内アーキテクチャの解読
（北村 朗、樺山 一哉）
ポスター討論
（奇数）
ポスター討論
（偶数）
撤去
若手の会会議
懇親会

■2019年9月26日（木）：年会3日目

階	部屋名	会場名	8	9	10	11	12	13
4F	天蘭	A会場		3CAA 最先端計測技術			男女共同参画・若手支援委員会企画シンポ	
	天玉	B会場		3SBA ヘム蛋白質の機能を司る構造・ダイナミクスとエネルギー流：理論と実験 （倭 剛久、David Leitner）			科研費説明会	
	天樹	C会場	ソーラボ2	3SCA 生体運動の多様性と普遍性 ―細胞内ダイナミクスから集団運動まで― （中村 修一、鹿毛 あずさ）			マルバーン・パナリティカル事業部（スペクトリス株式会社）	
	天葉	D会場	クロマ	3SDA 生命現象の理解を目指す光遺伝学の新展開 （角田 聡、井上 圭一）			サーモフィッシャーサイエンティフィックグループ 日本エフイー・アイ㈱	
	クリスタルルーム	E会場		3SEA 温度を基軸とした生物物理現象の理解 （原田 慶恵、岡部 弘基）			日本生物物理学会主催、先端バイオイメージング支援プラットフォーム 共催 イメージコンテスト	
	マーブルルーム	F会場		3SFA ナノ空間の生物物理 （多田隈 尚史、北川 大樹）			日本カンタム・デザイン株式会社	
	アイボリールーム	G会場		3SGA 超解像顕微鏡による生物物理学的生理学・病理学 （角山 貴昭、笠井 倫志）			ヤマト科学株式会社	
	アンバールーム	H会場		3SHA タンパク質の“質”を評価する （小川 覚之、内橋 貴之）			ブルカージャパン株式会社	
	天瑞・ホワイエ	PA会場/PB会場	貼付	ポスター掲示				ポスター討論（奇数）
	天瑞	展示会場		機器・試薬・書籍展示				
2F	オーチャード南	J会場			企業との意見交換会行事		リバネス	


14
15
16
17
18
19
20
21
ポスター討論
（偶数）
閉会式
ポスター撤去

参加者へのご案内

1. 年会受付と参加登録

◇年会受付

場　　所：2F ホワイエ

受付時間：8:00 - 17:00（9 月 26 日（木）は 14:00 まで）

◆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，領収証および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冊子をお受け取りください。

※参加証ホルダーは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：参加受付，展示会場など

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，参加登録費は免除されます。

◆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

		当日参加費	当日懇親会費
会員	正会員	￥12,000	￥8,000
	学生会員 / シニア会員	￥3,500	￥5,000
	学部学生	0	￥3,000
関連学会会員※	一般	￥12,000	￥8,000
	学生会員	￥3,500	￥5,000
非会員	一般	￥15,000	￥8,000
	大学院生	￥5,000	￥5,000
	学部学生	0	￥3,000

※関連学会とは，「オーストラリア生物物理学会（ASB）」，「日本物理学会」，「日本顕微鏡学会」です。

・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。
・若手奨励賞招待講演者，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。

◇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20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および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/57th_proceedings.pdf
ダウンロード ID：ambsj57
パスワード：miyazaki2019

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および J-STAGE にて予稿集の pdf ファイルが公開されます。

○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）

○ J-STAGE の生物物理のページ（https://www.jstage.jst.go.jp/browse/biophys/-char/ja）

◇プログラム検索（ウェブ版）【8月19日（月）公開予定】

年会ホームページで「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 月中旬公開予定】

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。

◇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：2F アトリウム

利用時間：8:00 - 18:30（9 月 26 日（木）のみ 15:30 まで）

※ 貴重品, 傘, コンピュータなどについては, 破損, 紛失などの責任を負いかねますので, お預かりできません。

※懇親会にご参加の際は，荷物をお引き取りください。

◇昼食

バイオフィジックスセミナー（1 - 3 日目），キャリア支援説明会（1 日目），男女共同参画・若手支援シンポジウム（3 日目），科研費説明会（3 日目）でお弁当とお茶が無料で提供されます。お弁当の配布方法については，20 ページのバイオフィジックスセミナーをご確認ください。

◇駐車場

シーガイアエリア内の各施設すべてに無料駐車場がありますので，ご自由にご利用いた

だけます。詳細につきましては，シーガイアの HP をご覧いただきますようお願いいたします。

◇ドリンクコーナー

展示会場内に，ドリンクコーナーを設けております。ドリンクコーナーでは，焼酎，コーヒー，ソフトクリームを取り揃えております。

◇インターネット

A 会場（天蘭）とポスター・展示会場（天瑞）では Wi-Fi をご利用いただけます。

◇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・総会シンポジウム

一般社団法人日本生物物理学会第 6 回会員総会を，年会 2 日目の 9 月 25 日（水）12:30 - 13:50 に A 会場（天蘭）で開催いたしますのでご出席ください。また，総会シンポジウムも開催します。

◇若手奨励賞招待講演

日本生物物理学会若手奨励賞及び若手招待講演賞の選考会である講演会（若手招待講演シンポジウム）を，年会 1 日目の 9 月 24 日（火）8:30 - 11:10 に A 会場（天蘭）で開催します。なお，授賞式は，3 日目のポスター発表終了後にポスター会場（ホワイエ）で行います。

◇学生発表賞

今大会の一般発表はポスター発表となりますので，ポスター発表で審査を行います。授賞式は，3 日目のポスター発表終了後にポスター会場（4F ホワイエ）で行います。

◇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 日目の 9 月 24 日（火）12:40 - 13:20 に A 会場（天蘭）で開催します。

◇分野別専門委員会

日　時：9 月 25 日（水）11:30 - 12:20
会　場：A 会場（天蘭）
対　象：分野別専門委員

昼　食：委員の皆様にはお弁当とお茶を提供します（整理券なし）。

◇懇親会

日　時：9 月 25 日（水）19:00 - 21:30
会　場：シーガイア 松泉宮ガーデン（屋外）
※当日参加も受け付けします（受付場所：総合受付デスク，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 26 日（木）11:30 - 12:40
会　場：A 会場（天蘭）
昼　食:お弁当とお茶が無料で提供されます（お弁当の配布方法については「バイオフィジックスセミナー」の項をご参照ください）。

◇キャリア支援説明会

日　時：9 月 24 日（火）11:30 - 15:00, 9 月 25 日（水）9:00 - 19:00
会　場：I 会場（2F オーチャード北）
対　象：就職を考えておられる学生や研究者など ※詳細は 49 ページをご参照下さい。
昼　食：9 月 24 日のみ，お弁当とお茶が無料で提供されます（学生・院生を優先して整理券を配布いたします。下記「バイオフィジックスセミナー」の項を参照）。
※下記の時間帯は個別相談を行っております。
9 月 24 日（火）16:00 - 19:00

◇科研費説明会

日　時：9 月 26 日（木）11:30 - 12:30
会　場：B 会場（天玉）
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。「バイオフィジックスセミナー」の項をご参照ください）。

◇バイオフィジックスセミナー

昼食を摂りながらの共催企業によるセミナーにご参加ください。なお，バイオフィジックスセミナーに参加するにはチケット（事前予約者）もしくは，当日配布する整理券が必要となります。当日整理券の配布については下記をご覧ください。

◆当日チケットの発券について

当日チケットについては下記のとおりに配布いたします。
時　間：9 月 24 日（火）- 26 日（木）8:00 -
場　所：PA 会場（展示受付）
※チケットは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。

◆チケットの注意事項

チケットは各日，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，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，チケットは無効となりお弁当はチケットをお持ちでない参加者に提供されますことをご了承ください。

◆バイオフィジックスセミナー受講時のお願い

バイオフィジックスセミナーは企業，団体等の共催によるセミナーです。参加される場合は最後までご聴講ください。また，共催者のアンケートには所属・氏名を記載して回答くださるよう，ご協力をお願いします。

◇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 4F 天瑞で行います。

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。一部，理事会・組織委員会等の承認を得て，録画を行う場合があります。

◇喫煙･飲食

喫煙は所定の喫煙場所でお願いいたします。講演会場内での飲食はバイオフィジックスセミナー，キャリア支援説明会，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となっております。

◇携帯電話

講演会場内での携帯電話による通話を禁止します。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

◇問い合わせ先

◆年会事務局

〒 532-0003 大阪市淀川区宮原 2-14-14 新大阪グランドビル 6F
株式会社エー・イー企画 内
Tel: 06-6350-7163　FAX: 06-6350-7164
E-mail: jbp2019@aeplan.co.jp（年会全般）

E-mail: e_jbp57@aeplan.co.jp（広告・展示関連）

◆参加登録･演題登録 システムサポートデスク

〒 602-8048 京都市上京区下立売通小川東入る 中西印刷株式会社内

E-mail: bsj2019sys-sprt@e-naf.jp

◆年会本部（会期中のみ） Tel: 080-4137-9158

座長, 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です。

◇座長へ

◆シンポジウム座長へのご案内

受　付：セッション開始の 10 分前までに各会場内の座長席近くの「タイムキーパ席」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，円滑な進行にご協力ください。事前に事務局に連絡した各講演者の講演時間等に変更が生じた場合は，タイムキーパーにお伝えください。

◆カレントトピックセッションの座長へのご案内

受　付：セッション開始の 10 分前までに各会場内の座長席近くの「タイムキーパ席」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：発表者の発表時間を厳守し，円滑な進行にご協力ください。

◇シンポジウム･カレントトッピクセッション発表者へ

◆発表方法

パソコンによる発表となります。必ずご自身のノートパソコンをお持ちください（会場にはパソコンを用意しておりません）。パソコンの操作は発表者ご自身で行ってください。発表データの作成や当日発表の際には以下の点にご留意ください。

注意 1）音声出力には対応しておりません。

注意 2）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してください。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ります。

注意 3）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 4）**切り替え装置に繋がるパソコンの映像出力端子は「HDMI」のみです。端子の形状が異なる場合は変換アダプターをお持ちください。**

注意 5）念のため発表に使用するパワーポイントファイルが入った USB メモリーをお持ちください。

注意 6）バッテリー切れに備え，電源アダプターをお持ちください。

注意 7）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，あらかじめ設定しておいてください。

◆シンポジウム発表者へ

受　　付：セッション開始の 15 分前までに会場の「PC 接続席」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 PC 切り替え装置に接続いたします。

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。若手招待講演シンポジウムの講演時間は，発表 10 分，討論 3 分，パソコンの交換に 2 分です。

◆カレントトピックセッションの発表者へ

受　　付：セッション開始の 15 分前までに会場の「PC 接続席」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 PC 切り替え装置に接続いたします。

講演時間：発表 20 分，質疑応答 10 分

◇ポスター発表者へ

◆ポスター発表要項

ポスターの貼付・展示，説明・討論，撤去：

		9 月 24 日（火）	9 月 25 日（水）	9 月 26 日（木）
貼付		8:00-8:30	8:00-8:30	8:00-8:30
説明・討論	奇数番号	16:30-17:30	16:50-17:50	12:50-13:50
	偶数番号	17:30-18:30	17:50-18:50	13:50-14:50
撤去		18:30-18:45	18:50-19:00	15:20-15:30

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。
4. 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），若手招待講演シンポジウム（Early Career Award in Biophysics Candidate Presentation Symposium），カレントトッピクセッション（Current Topic Sessions），ポスター発表（Poster Presentation）があります。

〈シンポジウム発表：（例）1SAA-03の場合〉

1（1 日目）S（シンポジウム）A（A 会場）A（AM: 午前）-03（発表順番）

〈若手招待講演シンポジウム：（例）1YK1045の場合〉

1（1 日目）Y（若手招待講演シンポジウム）K（K 会場）1045（10:45 講演開始）

〈カレントトッピクセッション（例）2CAP-01の場合〉

2（2 日目）C（カレントトピックセッション）A（A 会場）P（PM: 午後）-01（発表順番）

〈ポスター発表：（例）3Pos001の場合〉

3（3 日目）Pos（ポスター）001（ポスターパネル番号）

※学生発表賞に応募した発表には演題番号の右側に「*」がついております。

会員総会, 各種委員会のご案内

1. 一般社団法人日本生物物理学会第6回会員総会開催通知

一般社団法人日本生物物理学会第 6 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。

会員の皆様は，是非ご出席ください。

日　時：9 月 25 日（水）12:30 - 13:50

会　場：A 会場（天蘭）

議　長：会長 原田 慶恵

総会議題：

（1）報告事項

・平成 30 年度決算報告ならびに監査結果報告

・令和元 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

（2）第 6 回会員総会シンポジウム：「今後の生物物理学―50 年先への千里眼」

※詳細は 46 ページをご覧ください。

2. 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
ウェブサイト編集委員会	9 月 23 日（月・祝）	14:00-16:00	2F オーチャード南
生物物理編集委員会		16:30-18:30	
Biophysics and Physicobiology 編集委員会	9 月 24 日（火）	9:40-10:40	2F オーチャード南
出版委員会		10:40-11:40	
2019 年度第 2 回理事会（旧運営委員会）		11:40-12:30	
若手奨励賞選考委員会		16:10-17:10	
分野別専門委員会	9 月 25 日（水）	11:30-12:20	A 会場（天蘭）
会員総会・総会シンポジウム		12:30-13:50	
若手の会会議		16:00-17:00	2F オーチャード南
企業との意見交換会	9 月 26 日（木）	9:30-11:00	2F オーチャード南

※（）は法人化前の名前

謝　辞

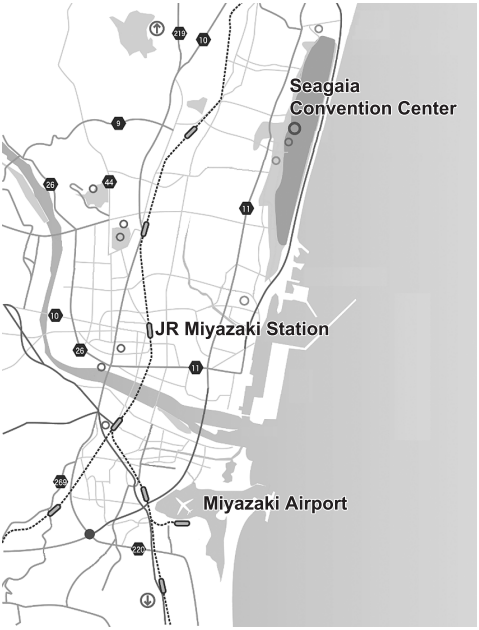
本年会の開催・運営にあたり，以下の団体よりご協力・ご援助いただきました。関係者一同より御礼を申し上げます。

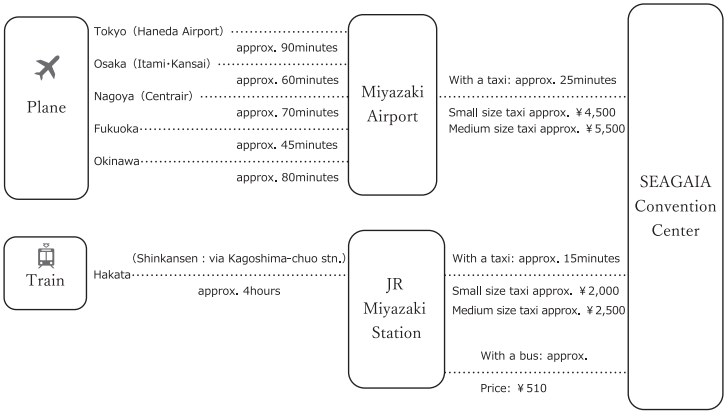
助成（敬称略）
（公財）宮崎県観光協会

共催／協賛（敬称略）
JST さきがけ「量子技術を適用した生命科学基盤の創出」
JST さきがけ「1 細胞解析」
JST さきがけ「生命機能メカニズム解明のための光操作技術」
新学術領域研究「代謝統合オミクス」
新学術領域研究「進化の制約と方向性」
新学術領域研究「遺伝子制御の基盤となるクロマチンポテンシャル」
新学術領域研究「発動分子科学：エネルギー変換が拓く自律的機能の設計」
新学術領域研究「分子夾雑の生命化学」
新学術領域研究「シンギュラリティ生物学」
新学術領域研究「温度生物学」
新学術領域研究「共鳴誘導で革新するバイオイメージング」

第 57 回日本生物物理学会年会
年会長　永井 健治

Access Information

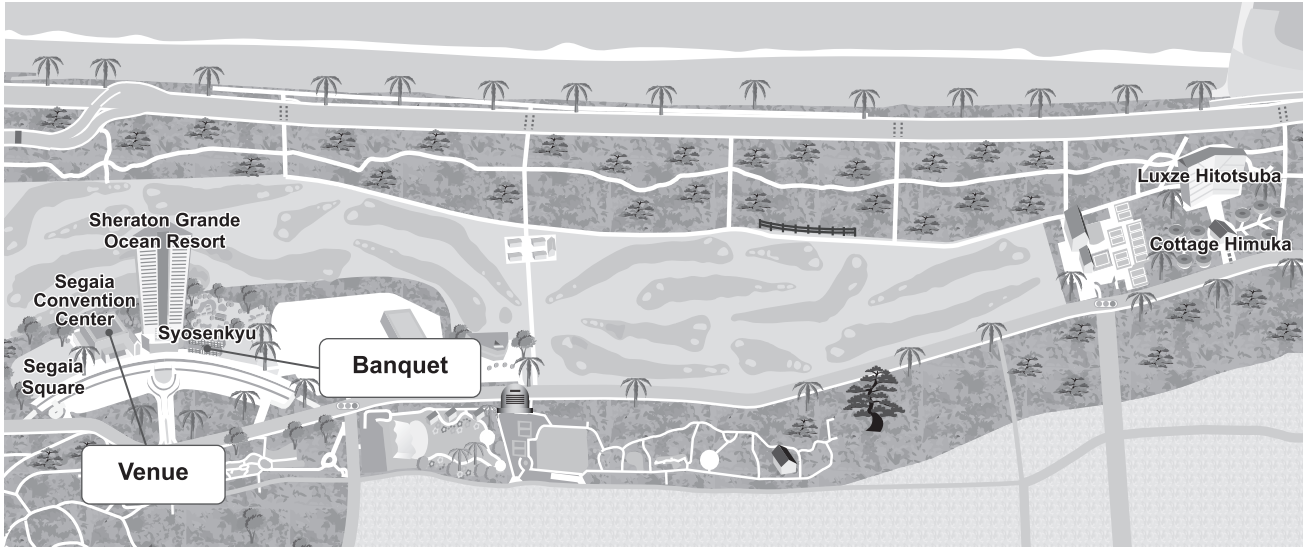




* Shared taxis are available from Miyazaki Airport to Seagaia Convention Center. Please come to the SHARED TAXI DESK at the airport for exclusive use of the 57th Annual Meeting participants.

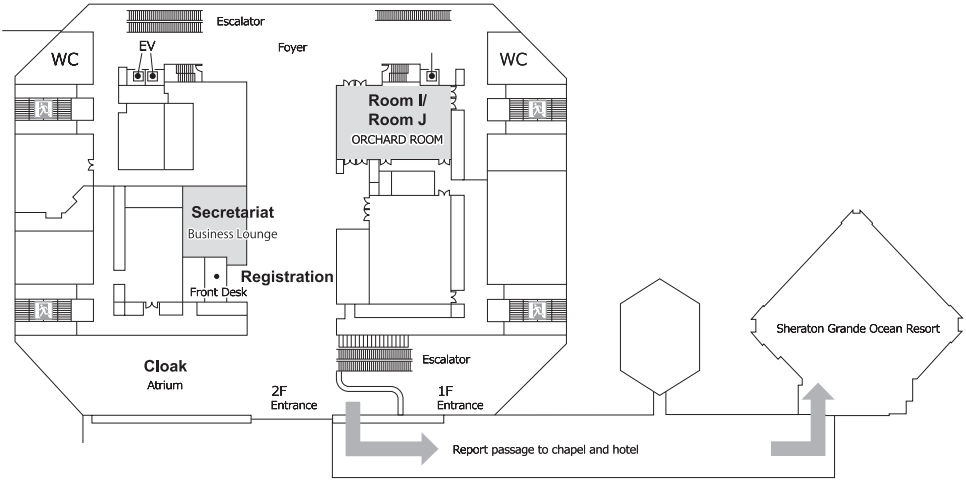
* Chartered buses will run on the meeting days.
Morning: JR Miyazaki Station → Seagaia Convention Center
After the meeting: Seagaia Convention Center → JR Miyazaki Station

* On the final day of the Meeting, chartered buses will run from Seagaia Convention Center to MIyazaki Airport after the closing remark.

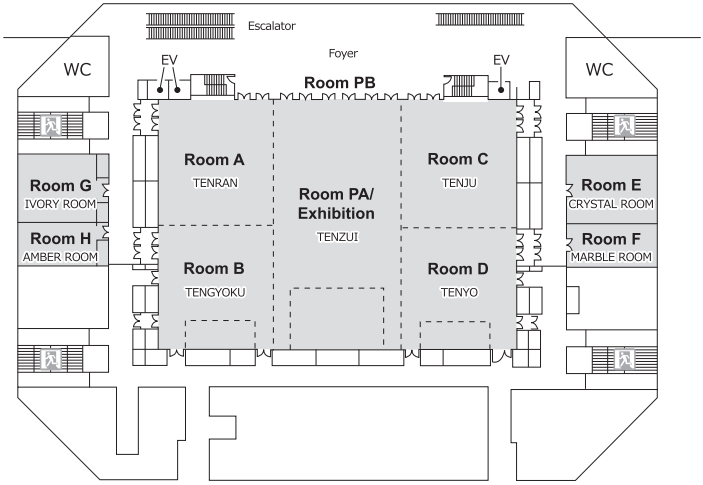


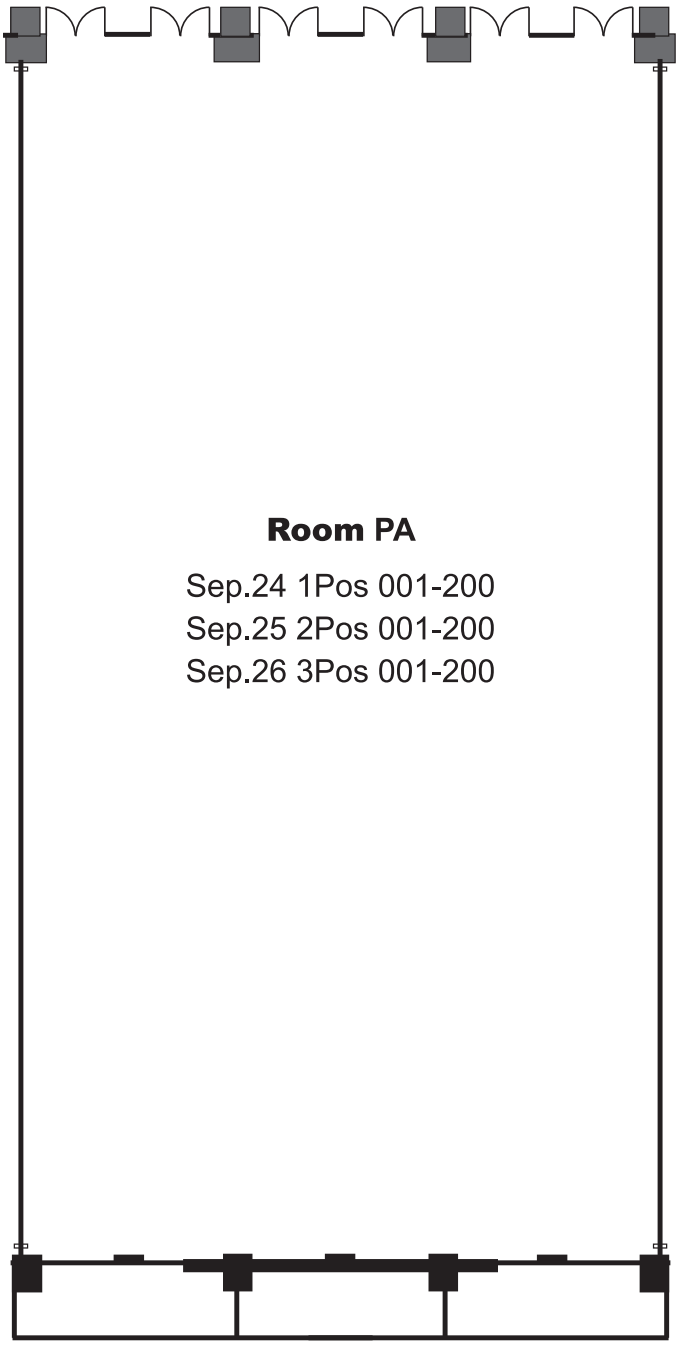
* Please note that it takes about 3km from Luxze Hitotsuba & Cottage Himuka to the Meeting Venue : Seagaia Convention Center.
Free Shuttle bus in Seagaia are available.

2nd Floor



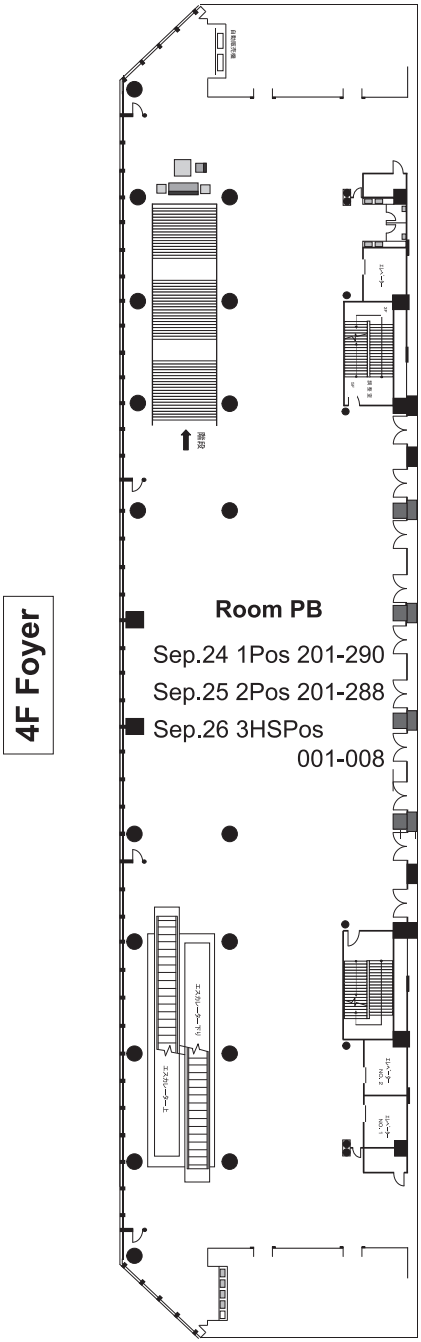
4th Floor





Room PA

Sep.24 1Pos 001-200
Sep.25 2Pos 001-200
Sep.26 3Pos 001-200


4F Foyer
Room PB
Sep.24 1Pos 201-290
Sep.25 2Pos 201-288
Sep.26 3HSPos
001-008

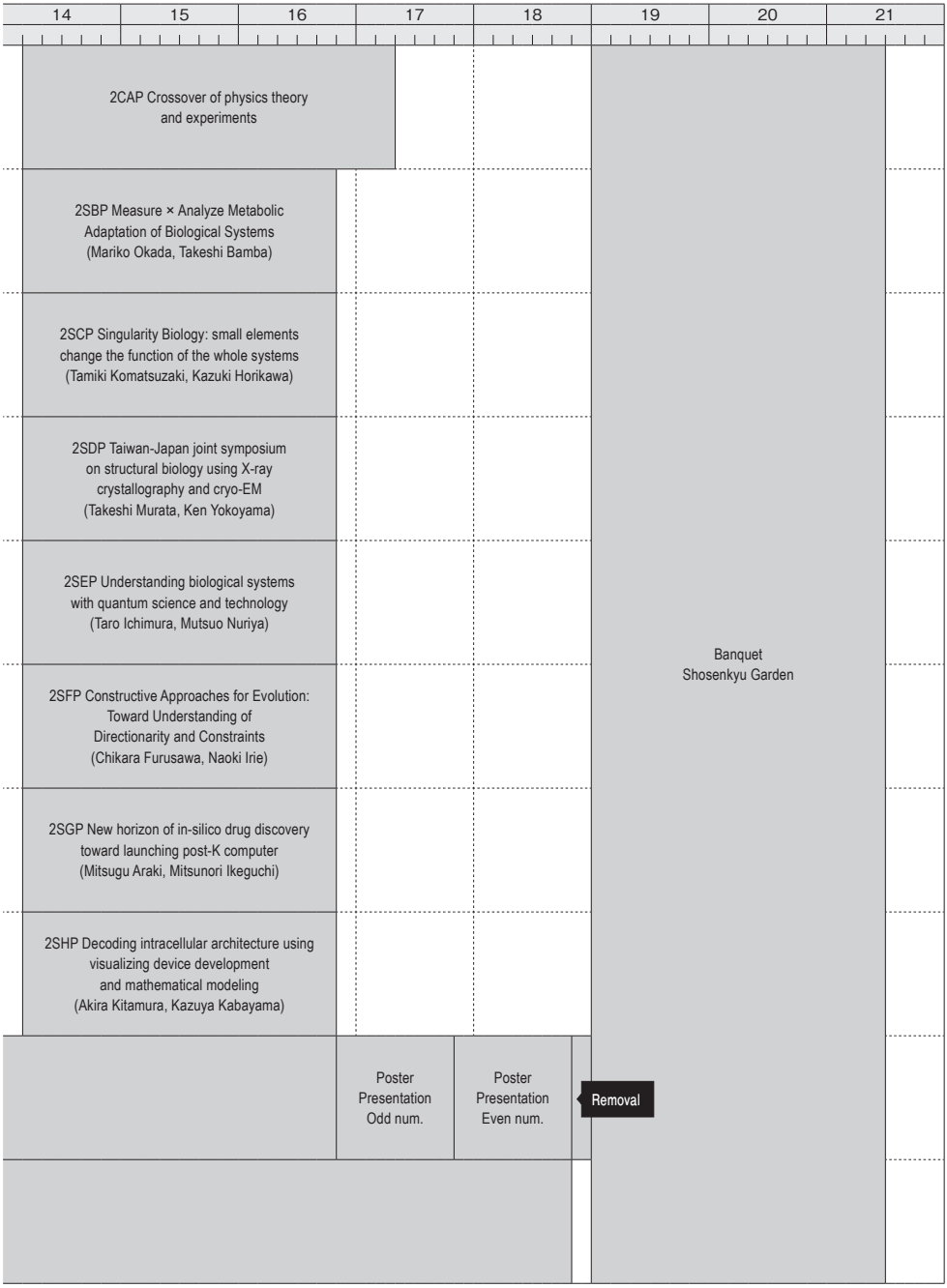
■ September 24 (Tue.) Day 1

Floor	Room	Venu	8	9	10	11	12	13
4F	TENRAN	Room A		1YA Early Career Award in Biophysics Candidate Presentation Symposium		Thorlabs Japan Inc.	Award Seminar of Outstanding Biophysics and Physicobiology Paper	
	TENGYOKU	Room B	YODAKA	1SBA Integrative approaches towards understanding of gene expression (Takaharu Mori, Shun-ichi Sekine)		NIKON INSTECH CO., LTD.		
	TENJU	Room C	Nanion	1SCA Utilization of soft compartments/ interfaces from nano to macroscale: Exploring the potential of living systems (Yusuke Sato, Masamune Morita, Yuki Suzuki)		Opto-Line, Inc.		
	TENYO	Room D	Spectris	1SDA What can or cannot do by cryo-EM? The forefront of Structural Life Science (Masahide Kikkawa, Atsushi Nakagawa)		HAMAMATSU PHOTONICS K.K.		
	CRYSTAL ROOM	Room E		1SEA Physics of chromatin dynamics – towards understanding the regulation of gene expression (Yuma Ito, Akatsuki Kimura)		Nihon Waters K.K.		
	MARBLE ROOM	Room F		1SFA Toward "Ari-No-Mama" visualization to reveal biological functions–Resonance between life science and optical technology– (Atsushi Miyawaki, Tomomi Nemoto)		YODAKA CO., Ltd.		
	IVORY ROOM	Room G	Thorlabs	1SGA Frontiers in multi-scale mechanobiology of muscle and vascular system (Mitsuhiro Iwaki, Yuji Hara)		Protein Data Bank Japan (PDBj)		
	AMBER ROOM	Room H	INTEGRA	1SHA Control of biological functions with hydrostatic pressure stimulation (Hiroaki Hata, Masayoshi Nishiyama)		DKSH Japan		
	TENZUI・FOYER	Room PA/ Room PB	Set Up	Poster Display				
	TNNZUI	Exhibition			Exhibition			

14	15	16	17	18	19	20	21
1CAP Protein: Structure･function･design							
1SBP ASB-BSJ Joint Symposium: Current challenges in biophysics centering on biomolecular interactions and the underlying forces (Takayuki Nishizaka, Marc Kvansakul)							
1SCP Cutting-edge brain research from a biophysical perspective (Takashi Tominaga, Bernd Kuhn)							
1SDP Current status and issues of protein solution biophysics (Sususmu Uchiyama, Saeko Yanaka)							
1SEP What is “Single-cell PRESTO” doing? (Katsuyuki Shiroguchi, Madoka Suzuki)							
1SFP Toward the chemical reaction control in biological environment by high-sensitive hydrogen detection (Ichiro Tanaka, Hisroshi Ishikita)							
1SGP New horizon in molecular observation through high-dimensional data-driven and measurement informatics approaches (Takanori Kigawa, Yasuhiro Matsunaga)							
1SHP Frontier of structure-function studies to unveil diverse GPCR signaling (Kota Katayama, Ryoji Suno)							
		Poster Presentation Odd num.	Poster Presentation Even num.	Removal			

■ September 25 (Wed.) Day 2

Floor	Room	Venu	8	9	10	11	12	13
4F	TENRAN	Room A	Tokyoinst →	2CAA Biophysics of cell biology		Experts Committee	General Assembly Symposium	
	TENGYOKU	Room B		2SBA Invitation to multimolecular crowding (Kazuhito Tabata, Daisuke Miyoshi)		Olympus Corporation		
	TENJU	Room C		2SCA Challenges of bioinformatics for the era of molecular structure big-data (Tsuyoshi Shirai, Tohru Terada)		Malvern Panalytical division of Spectris Co., Ltd.		
	TENYO	Room D		2SDA Nonequilibrium Energetics of Biological Molecular Machines (Chun-Biu Li, Shoichi Toyabe)		Thermo Fisher Scientific K.K.		
	CRYSTAL ROOM	Room E		2SEA Frontiers of Synchrotron Radiation Biophysics (Hiroyuki Iwamoto, Hiroshi Sekiguchi)		OptoSigma		
	MARBLE ROOM	Room F		2SFA Elucidation of biological functions by optical control (Yoshinori Shichida, Hisao Tsukamoto)		Anton Paar Japan K.K.		
	IVORY ROOM	Room G		2SGA How is 'ENERGY' generated/transferred across the cellular systems? (Kei Wada, Yoichi Sakakibara)		Beckman Coulter K.K.		
	AMBER ROOM	Room H	Quantum →	2SHA Challenges to get insight into unsolved problems of dynamic response in proteins (Yu Takano, Yasushige Yonezawa)		SCIEX		
	TENZUI・FOYER	Room PA/ Room PB	Set Up	Poster Display				
	TNNZUI	Exhibition			Exhibition			


14
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16
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21
2CAP Crossover of physics theory and experiments
2SBP Measure × Analyze Metabolic Adaptation of Biological Systems (Mariko Okada, Takeshi Bamba)
2SCP Singularity Biology: small elements change the function of the whole systems (Tamiki Komatsuzaki, Kazuki Horikawa)
2SDP Taiwan-Japan joint symposium on structural biology using X-ray crystallography and cryo-EM (Takeshi Murata, Ken Yokoyama)
2SEP Understanding biological systems with quantum science and technology (Taro Ichimura, Mutsuo Nuriya)
2SFP Constructive Approaches for Evolution: Toward Understanding of Directionarity and Constraints (Chikara Furusawa, Naoki Irie)
2SGP New horizon of in-silico drug discovery toward launching post-K computer (Mitsugu Araki, Mitsunori Ikeguchi)
2SHP Decoding intracellular architecture using visualizing device development and mathematical modeling (Akira Kitamura, Kazuya Kabayama)
Poster Presentation Odd num.
Poster Presentation Even num.
Removal
Banquet
Shosenkyu Garden

■ September 26 (Thu.) Day 3

Floor	Room	Venu	8	9	10	11	12	13
4F	TENRAN	Room A		3CAA State of the art measurement technologies			Gender Equality & Young Researchers Support Symposium	
	TENGYOKU	Room B		3SBA Structure, Dynamics and Energy Flow that Govern Heme Protein Functions: Theory and Experiments (Takahisa Yamato, David Leitner)			KAKENHI Guide Meeting	
	TENJU	Room C	Thorlabs2	3SCA Diversity and universality of motile mechanism of living things: From intracellular dynamics to collective motion (Shuichi Nakamura, Azusa Kage)			Malvern Panalytical division of Spectris Co., Ltd.	
	TENYO	Room D	Chroma	3SDA Optogenetics: Applying photoreceptor for understanding biological phenomena (Satoshi Tsunoda, Keiichi Inoue)			FEI Company, part of Thermo Fischer Scientific	
	CRYSTAL ROOM	Room E		3SEA Single-Cell Thermal Biology (Yoshie Harada, Kohki Okabe)			Advanced Bioimaging Support (ABiS)	
	MARBLE ROOM	Room F		3SFA Biophysics in Nano-space (Hisashi Tadakuma, Daiju Kitagawa)			Quantum Design Japan, Inc.	
	IVORY ROOM	Room G		3SGA Biophysical Physiology and Pathology by the Application of Superresolution Microscopy (Taka A. Tsunoyama, Rinshi S. Kasai)			Yamato Scientific Co., Ltd.	
	AMBER ROOM	Room H		3SHA The Quality of Proteins - Multiple Approaches for Protein Evaluation - (Tadayuki Ogawa, Takayuki Uchihashi)			Bruker Japan K.K.	
	TENZUI・FOYER	Room PA/ Room PB	Set Up	Poster Display				Poster Presentation Odd num.
	TNNZUI	Exhibition		Exhibition				
2F	ORCHARD South	Room J					Leave a Nest Co., Ltd	

14	15	16	17	18	19	20	21
Poster Presentation Even num.	Closing Ceremony	Removal					

Information for Participants

1. Registration

◇Registration desk

Location：2F Foyer

Open Hours：8:00 - 17:00 [8:00 - 14:00 on Sep. 26 (Thu.)]

◆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 and near the building entrance.)

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.

◆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

		Registration fee	Banquet fee
Member	Regular	￥12,000	￥8,000
	Student / Senior	￥3,500	￥5,000
	Undergraduate Student	free	￥3,000
Related Society Member*	Regular	￥12,000	￥8,000
	Student	￥3,500	￥5,000
Non-Member	Regular	￥15,000	￥8,000
	Student	￥5,000	￥5,000
	Undergraduate Student	free	￥3,000

*The Physical Society of Japan, the Japanese Society of Microscopy, or the Australian Society for Biophysics

・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.
・The "Early Career Award in Biophysics" Candidate presenters, the Biophysics and Physicobiology
Outstanding Paper Award presenter and the representative of Biophysics and Physicobiology Editors' Choice awardee 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.

◇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. 20 (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/57th_proceedings.pdf
Download ID: ambsj57
PW: miyazaki2019

The program (presentation title, presenter's name and affiliation) and the online abstracts will be released on the BSJ2019 web site. A half year later after the meeting, the abstracts will be posted on the J-Stage web site which is linked from the BSJ web site.
BSJ web site: http://www.biophys.jp/ann/ann02.html
J-Stage web site: http://www.jstage.jst.go.jp/browse/biophys/-char/ja

◇Program search system (Web version)【Release date: Aug. 19 (Mon.)】

Program search system will be released on the BSJ2019 web site.

◇Free app to search and browse Program & Abstracts【Release date: Mid-Sep】

A free app for smart phones (iPhone /Android) and tablets (iPad /iPod Touch /Android) can be downloaded from App Store or Google Play.

◇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: 2F Atrium

Open Hours: 8:00 - 19:00 [8:00 - 15:30 on Sep. 26 (Thu.)]

Cloakroom is available as follows. No valuable, computer, or umbrella can be checked in to the cloak since the society/meeting does not hold any responsibility for loss or damage of your items.

◇Lunch

Free lunch:

Free lunch will be provided at biophysics seminars (day 1-3), Career Support Meeting (day 1), Gender Equality & Young Researchers Support Symposium (day 3), and KAKENHI Guide Meeting (day 3). Lunch tickets will be distributed in the morning of day 1-3. Please refer to biophysics seminar page.

◇Paging service

No paging service is available to call an individual except for an emergency.

◇Parking

Free private parking is available on site (reservation is not needed).

◇Drink Service

A drink corner is available in the exhibition hall. We have Shochu; a traditional Japanese distilled spirit, coffee and soft ice cream there.

◇Internet Access

Wi-Fi networks of Room A（TENRAN）and Room PA（TENZUI）is available.

3. Programs & Events

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

◇“Early Career Award in Biophysics” Candidate presentations

Date & Time: Sep. 24 (Tue.) 8:30 - 11:10

Place: Room A (TENRAN)

◇Student Presentation Award

In this year's meeting, the awardees will be selected from the applicants who are presenting poster presentations.

The names of the awardees will be released at the meeting Award ceremony on the last day of the annual meeting.

◇Banquet

Date & Time: Sep. 25 (Wed.) 19:00 - 21:30

Place: Seagaia Shosenkyu Garden

*On-site registration is available at the registration desk or the banquet reception desk.

*There is no shuttle bus service and we'll head to the venue on foot (Those who need any assistance, please do not hesitate to consult with staff at the general information).

◇Biophysics Seminar

Come and join the biophysics seminar hosted by the co-sponsored companies. Lunch tickets will be distributed at the biophysics seminar desk as shown below. Onsite tickets are also available. Please see below for the details.

◆Distribution of lunch tickets

Biophysics seminar desk

Hours: 8:00 -

*The desk will be closed when all the tickets are distributed.

Location: PA room (TENZUI)

*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 biophysics 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.

◆Request

We kindly ask you to attend till the last. Also, please enter your affiliation and name in the questionnaire provided at the biophysics seminar as possible as you can.

◇Exhibition

Instruments, reagents, software, books, etc. are displayed at the exhibition hall (ENZUI).

4. Prohibited Items

◇Photography & recording

Photography and recording with camera, video, mobile phone and any device is NOT allowed at the meeting site. Please note that some recording may be performed after obtaining the board meeting's approval.

◇Smoking

Please smoke in a designated area.

◇Mobile 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: 080-4137-9158 *phone number reachable during the meeting)

◇Before or after the meeting

◆BSJ2019 Secretariat

A & E planning Co., Ltd.
Shin-Osaka Grand Bldg. 6F, 2-14-14, Miyahara, Yodogawa-ku, Osaka 532-0003
Tel: 06-6350-7163 / Fax: 06-6350-7164
Support team: jbp2019@aeplan.co.jp
Exhibition and Advertisement secretariat: e_jbp57@aeplan.co.jp

◆Registration and abstract submission support desk

Nakanishi Printing Company
Ogawa-higashiiru, Shimodachiuri-dori, Kamigyo-ku, Kyoto 602-8048
bsj2019sys-sprt@e-naf.jp

Information for Chairpersons and Presenters

◇Language

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

◇For Chairpersons of symposia

All chairpersons are requested to be seated 10 minutes before the starting time. Please inform the staff standing by near you if any change from the schedule is necessary. The staff will assist with timing. Please make any necessary arrangements regarding bell signals before the session.

◇For speakers of symposia and current topic sessions

◆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 × 768). The higher resolutions would possibly lose some information.

3) Our staff will connect your laptop to a switcher.
4) **The output connector of your laptop should be "HDMI". 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 Speakers of Symposia

Please come to the "PC Preview Desk" in the assigned room by 15 minutes before the start of the session. Our staff will connect your computer to a switcher.
Time allocation: Symposium: Time allocation will be controlled by chairpersons. "Early Career Award in Biophysics" Candidate Presentation: Presentation 10 minutes + Discussion 3 minutes + Laptop change 2 minutes.

◆For speakers of current topic sessions

Please come to the "PC Preview Desk" in the assigned room by 15 min before your presentation. Our staff will connect your laptop to a switcher.
Time allocation: Presentation 20 minutes + Discussion 10 minutes

◇For poster presenters

		Day 1, Sep. 24 (Tue.)	Day 2, Sep. 25 (Wed.)	Day 3, Sep. 25 (Thu.)
Setup		8:00-8:30	8:00-8:30	8:00-8:30
Presentation / Discussion	Odd Numbers	16:30-17:30	16:50-17:50	12:50-13:50
	Even Numbers	17:30-18:30	17:50-18:50	13:50-14:50
Removal		18:30-18:45	18:50-19:00	15:20-15:30

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

<Speech at symposium: (Ex.) 1SAA-03>

Presentation day (1, Sep. 24) + Symposium (S) + Session room (Room A) + AM (A) + Order of the talk (No.3)

<“Early Career Award in Biophysics” Candidate Symposium: (Ex.) 1YK1045>

Presentation day (1, Sep. 24) + Young Scientists (Y) + Session room (Room K) + Starting time of the talk (10:45)

<Speech at Current Topics Session : (Ex.) 2CAP-01>

Presentation day (2, Sep. 25) + Current Topics Session (C) + Session room (Room A) + PM (P) + Order of the talk (No.3)

<Poster presentations: (Ex.) 1Pos001>

Presentation day (1, Sep. 24) + Poster (Pos) + Panel number (001)

◆ For presentations which are eligible for consideration for Student Presentation Award, “*” is attached on the right side of a presentation number.

ABiS イメージングコンテスト 2019

日　時：9 月 26 日（木）11:30 - 12:30
会　場：E 会場（クリスタルルーム）
司　会：宮田 真人（大阪市立大学），藤井 律子（大阪市立大学）
共　催：新学術領域研究・学術研究支援基盤形成「先端バイオイメージング支援プラットフォーム（ABiS）」
※バイオフィジックスセミナー形式で行います。

57 回年会企画として，「ABiS イメージングコンテスト」を開催いたします。本コンテストでは，生物物理学研究に基づくあらゆる画像（静止画，動画を問わない）を対象とし「美的部門」，「知的部門」，「ユーモア部門」の 3 つのカテゴリー毎に受賞者を決定します。
受賞者の選考方法はご応募いただきました画像，動画を日本生物物理学会年会の参加者による投票により 10 件ほど選出します。選出された 10 件については，バイオフィジックスセミナー・イメージコンテストにて投影と応募者による 3 分程度の説明の後，会場にいる聴衆全員による投票を行います。
各カテゴリーの受賞者は，同日の表彰式（9 月 26 日（木）14:50 - 15:20　ホワイエ）にて発表します。受賞者には賞状と副賞が授与されます。

第 6 回会員総会シンポジウム：今後の生物物理学―50 年先への千里眼

オーガナイザー：日本生物物理学会　理事会
日　時：9 月 25 日（水）12:30 ～ 13:50（会員総会中）
会　場：A 会場（サミットホール天蘭）
司　会：小松崎民樹

概要：大沢文夫氏を追悼するシンポジウムという位置づけで，大沢氏の門下生である郷氏と「大沢流手作り統計力学」の本の作成にご尽力された大沼氏にご講演いただいた後で，大沢氏がおっしゃっていた「生物物理学とは生物学としても物理学としても面白いテーマでなければいけない」という言葉の意味をみんなで考えてみたいと思います。生物物理学の特徴のひとつはその自由な発想のもとで展開してきた異分野融合力にあると思います。生物学と物理学だけでなく，生物学を軸とした色々な異分野融合は今後も強化されていくものと想像されます。パネリストの方々も交えて，情報熱力学，計測・分析科学，計算科学，ホジキン・ハックスリーのような応用数学など他分野へのフィードバックの可能性なども含めて，今後の 50 年を展望した自由な討論を行い，会員一同，大沢氏の感想，批評を想像したいと思います。

講演者・プログラム：

1. 郷　通子氏（元日本生物物理学会長，名古屋大学名誉教授，お茶の水大学名誉教授）
「高分子，統計力学，そして生物へ」

2. 大沼　清氏（長岡技術科学大学 技術科学イノベーション専攻／生物機能工学専攻）
「大沢流てづくり統計力学」から得た発生生物学のヒント

3. パネルディスカッション「生物学と異分野の融合の観点」
岡田康志氏（東京大学大学院理学系研究科 / 理化学研究所 BDR）情報熱力学，計測科学と生物学
冨樫祐一氏（広島大学院統合生命科学研究科 / 理化学研究所 BDR）数理科学と生物学
青野真士氏（慶應義塾大学環境情報学部 / 東京工業大学地球生命研究所）生物学と計算科学
根岸瑠美氏（東京大学定量生命科学研究所）生物学と分析科学
郷　通子氏　　物理学と進化学
大沼　清氏　　生物学と発生学

一般社団法人日本生物物理学会　第 8 回 Biophysics and Physicobiology 論文賞受賞講演会
The 8th Award Seminar for 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 24 日（火）12:40 ～ 13:20 ／ Sep. 24 Tue.
場　所：A 会場（サミットホール天蘭）／ Room A (Summit Hall Tenran)
形　式：講演会／ Lecture

第 8 回 Biophysics and Physicobiology 論文賞受賞者

David J. Castillo[1]，中村修一 [2]，森本雄祐 [3]，蔡栄淑 [1]，上池伸徳 [1]，工藤成史 [2]，南野徹 [1]，難波啓一 [1,4]

David J. Castillo[1], Shuichi Nakamura[2], Yusuke V. Morimoto[3], Yong-Suk Che[1], Nobunori Kami-ike[1], Seishi Kudo[2], Tohru Minamino[1] and Keiichi Namba[1,4].

[1] 阪大・院生命機能，[2] 東北大・院工，[3] 九工大・院情工，[4] 理研・BDR&SPring-8

[1]Grad. Sch. Frontier Biosci., Osaka Univ., [2]Grad. Sch. Eng., Tohoku Univ., [3]Grad. Sch. Comp. Sci. and Sys. Eng., Kyushu Inst. Tech., [4]RIKEN BDR & SPring-8

細菌べん毛モーター固定子の組み込みと活性化のメカニズム

Stator assembly and activation mechanism of the bacterial flagellar motor

The bacterial flagellar motor is a rotary nanomachine fueled by the transmembrane electrochemical potential of ions, i.e., ion motive force (IMF). The flagellar motor comprises a rotor and multiple stators. The stator proteins MotA and MotB form a transmembrane complex containing two proton channels, and each stator unit is anchored to the peptidoglycan (PG) layer via a PG binding (PGB) motif of MotB. The stator can control its ion conductivity by a “plug” that resides in the periplasmic region of MotB. The stator unit is believed to be activated and inactivated by the detachment and attachment of the plug segment from and to the proton channel, respectively. Although 10 stator units can be incorporated to the motor, they alternate in assembly and disassembly even during rotation, accompanying transition between active and inactive states. Stator assembly and activation depend on external load, external ion concentration, and IMF. Castillo and Nakamura et al. [1] analyzed the rotation of the mutant flagellar motor lacking residues 72-100 of MotB, corresponding to a linker between the transmembrane proton channel and PGB domains, and suggested its critical role for load-dependent stator assembly. This seminar will review the current knowledge on stator assembly dynamics, and will discuss the mechanism by which flagellar stators sense load and input energy.

1. Castillo DJ and Nakamura S *et al.* (2013) The C-terminal periplasmic domain of MotB is responsible for load-dependent control of the number of stators of the bacterial flagellar motor. *Biophysics* 9: 173–181.

男女共同参画・若手支援委員会企画シンポジウム
20 代，30 代を駆け抜けて：伝えたいこと，聞きたいこと

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 26 日（木）11:30 ～ 12:45（バイオフィジックスセミナーの時間帯）
会　場：A 会場（サミットホール天蘭）
言　語：日本語
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：プレゼンテーションとグループディスカッション
司　会：須藤雄気（岡山大学）
発表者：酒井佳寿美（京都大学），田中俊一（京都府立大学），寺川剛（京都大学）（当日変更可能性あり）

概　要：この数十年，大学院重点化や法人化などで学生を含めた若手研究者の環境は大きく変化し，博士課程入学者の増加（とその後の減少），任期付きポストの増加などがおこりました。また，社会環境の変化により，企業での研究環境も大きく変動しています。一般に，現場では 20 ～ 30 代の方々の活躍が重要ですが，結婚や子育てなど生活面において大きく変化する時期と重なるため，大学や社会状況が急速に変化する中で，自身の生活と研究者としてのキャリアアップをどう両立していくかが大きな課題となっています。

本企画シンポジウムでは，はじめに，30 代，40 代の多様なキャリアパスを持つ 3 名の発表者の方に，各人 15 分程度，ターニングポイント，重要だった選択など，研究者としてどのようにキャリアアップしてきたかをお話しいただきます。また，生活とどのように両立してきたかについてもお話いただきます。

酒井博士は，大学卒業後メーカーに勤務され，結婚を経て，大学でプロジェクト技術員として勤務した後，大学院で博士課程を取得され，現在ポスドクをされています。

田中博士は，博士課程を修了した後，ポスドクとして一年間研究，日本企業で二年間勤務，米国大学で四年間研究（当時所属企業から出向），米国企業で一年間勤務，その後アカデミアに戻り，現在は准教授として働かれています。2 男 1 女の 3 児のパパで，中間反抗期を迎えた長男と次男に悪戦苦闘中とのことです。

寺川博士は，博士課程を修了した後，海外でポスドクをされて，現在は助教をされています。また，夫婦ともに研究経験があり，1 児のパパになります。

3 名からのお話を拝聴した後，参加者全員で，興味ある議題に関してグループになり，グループディスカッションを行います。経験談や現在抱えている問題点などを共有することによって，今後のキャリアプランのなんらかの参考になればと考えています。学生，若手研究者の方の参加を歓迎します。また，ご自身のご経験をお話していただく，幅広い年齢の研究者の方の参加も歓迎いたします。部屋には，ジョイントマットスペースを用意しております。お子様連れの方も，お子様とお昼を食べる感覚でご参加いただければと存じます（お子様のお昼は各自ご持参下さい）。

キャリア支援説明会

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

日　時：9 月 24 日（火）第 1 部 11:30-12:15，第 2 部 13:00-13:45，第 3 部 14:00-14:45，個別相談会 16:00-19:00
9 月 25 日（水）第 4 部 9:00-9:45，第 5 部 10:00-10:45，第 6 部 11:00-11:45，個別相談会 12:00-17:00

会　場：2F オーチャード北

形　式：セミナー 第 1 部のみランチョンセミナー **説明はすべて日本語で行われます。**

概要:若手研究者や学生の今後のキャリア構築の一助となるように, 今年度も「キャリア支援説明会」を開催します。昨年の反響を受けて本年会は，(株) アカリクから吉野宏志氏を迎えて大学院生やポストドクター向けの就職支援活動セミナーを実施します。第 1 部のみランチョンセミナーのため参加者数に限りがありますが，第 2 部以降から参加することも可能です。また昨年度と同様に今年度も個別相談会を実施いたします。人前では難しい個人的な相談等があれば，是非活用ください。

プログラム：　　**9 月 24 日（火）**

第 1 部　11:30-12:15　博士・PD の方の為の就活ガイダンス
徐々に企業の採用対象として存在感を増してきている博士人材ですが，研究実績があっても状況の把握や伝え方を誤れば機会を逃してしまいます。そこで，①企業の採用活動の現在，②博士や PD の就職活動の実態，③博士人材が活躍できる企業の探し方を中心に紹介します。博士・PD 以外の方もご参加いただけます。

第 2 部　13:00-13:45　「専門外就職」へ向けて何をすべきか
専門性が高いほど技術や知識がそのまま活用できる仕事は少なくなりますが，その専門性を構成する要素を紐解くことで「専門外」の領域で幅広く活躍することが視野に入ってきます。専門外就職を目指す上で特に注意すべき「一貫性」を中心に書類の書き方や面接対策などのノウハウを解説します。

第 3 部　14:00-14:45　理系大学院生の就活ケーススタディ
周りを見れば大枠としてどのように就職活動しているか見えてくるかと思いますが，自身に最適な方法や目標を見つけるのは至難の業です。ここでは実際にアカリクで支援した方の情報をもとにして，状況に応じてどのような戦略や戦術が有効なのかを解説します。

個別相談会 16:00-19:00
就職活動・キャリアに関する悩みや不安を気軽にご相談ください。また就活ノウハウや企業での待遇面など分からないことがあれば遠慮なくお尋ねください。相談予約方法は 1 ～ 3 部のセミナー中に提示します。

9 月 25 日（水）

第 4 部　09:00-09:45　博士・PD の方の為の就活ガイダンス
徐々に企業の採用対象として存在感を増してきている博士人材ですが，研究実績があっても状況の把握や伝え方を誤れば機会を逃してしまいます。そこで，①企業の採用活動の現在，②博士や PD の就職活動の実態，③博士人材が活躍できる企業の探し方を中心に紹介します。博士・PD 以外の方もご参加いただけます。

第 5 部　10:00-10:45　「専門外就職」へ向けて何をすべきか
専門性が高いほど技術や知識がそのまま活用できる仕事は少なくなりますが，その専門性を構成する要素を紐解くことで「専門外」の領域で幅広く活躍することが視野に入ってきます。専門外就職を目指す上で特に注意すべき「一貫性」を中心に書類の書き方や面接対策などのノウハウを解説します。

第 6 部　11:00-11:45　理系大学院生の就活ケーススタディ
周りを見れば大枠としてどのように就職活動しているか見えてくるかと思いますが，自身に最適な方法や目標を見つけるのは至難の業です。ここでは実際にアカリクで支援した方の情報をもとにして，状況に応じてどのような戦略や戦術が有効なのかを解説します。

個別相談会：　12:00-17:00
就職活動・キャリアに関する悩みや不安を気軽にご相談ください。また就活ノウハウや企業での待遇面など分からないことがあれば遠慮なくお尋ねください。相談予約方法は 1 ～ 6 部のセミナー中に提示します。

「科研費」の最近の動向
Reorganization of KAKENHI: Current Activities of JSPS

世話人：原田慶恵（大阪大学蛋白質研究所，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：Yoshie Harada (Institute for Protein Research, Osaka University; Program Officer, Research Center for Science Systems, JSPS)

日　時：9 月 26 日（木）11:30–12:30（バイオフィジックスセミナーの時間帯）

会　場：B 会場（天玉）

昼　食：お弁当とお茶が無料で提供されます。

形　式：日本語による講演

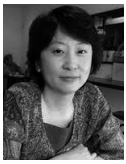
緊急！パネルディスカッション～地方創生と大学改革の接点～
Point of contact between Regional revitalization and University reforms

*「科研費」の最近の動向の講演後に続けて開催いたします。

パネリスト（順不同）：
原田慶恵（大阪大学蛋白質研究所・教授 ／ 学会長）
永井健治（大阪大学産業科学研究所・教授 ／ 年会長）
生田知子（文部科学省・高等教育局・大学改革官）
福田卿也（博報堂）
渡邉朋信（理化学研究所・チームリーダー）

司会：
樺山一哉（大阪大学理学研究科・准教授）






第 1 日目（9 月 24 日（火））／Day 1（Sep. 24 Tue.）

8:30〜11:10　A 会場／Room A：サミットホール天蘭／Summit Hall Tenran
1YA　日本生物物理学会若手奨励賞選考会
Early Research in Biophysics Award Candidate Presentations

オーガナイザー：男女共同参画・若手支援委員会
Organizer: Promotion of Gender Equality and Young Researchers Committee

Biophysical Society of Japan (BSJ) grants “Early Career Award in Biophysics” and “Early Career Presentation Award” to young BSJ members for their excellent presentations that show great potential to contribute to the progress of biophysics. In this 15th year, we received 27 highly qualified applications. After the first round of competitive screening based on submitted documents, the following ten applicants were selected as the young invited speakers. In this symposium, each speaker will make 10-minute presentation followed by 3-minute discussion as the second round of screening. Up to five awardees of the Early Career Award in Biophysics will be selected and announced at the banquet held in the evening of the second day. The Early Career Presentation Award will be given to the rest of the excellent invited speakers. We welcome all the BSJ members to attend this symposium to foresee the future of biophysics in Japan through the speakers and their researches.

8:30　**岡本　和子**　2Pos091
1YA0830　**転写因子の振る舞いとクロマチンのゆらぎの関係性を 1 分子計測によって解析する**
Single molecular dynamics of transcription factors are controlled by diffusion movement of chromatin
○岡本 和子 [1], 岡田 康志 [1], 阿倍 訓也 [2], 渡邉 朋信 [1]（[1]理化学研究所 生命機能科学研究センター, [2]理化学研究所 バイオリソース研究センター）
Kazuko Okamoto[1], Yasushi Okada[1], Kuniya Abe[2], Tomonobu M Watanabe[1] ([1]*RIKEN BDR*, [2]*RIKEN BRC*)

8:45　**佐藤　慎哉**　3Pos133
1YA0845　**リガンド非結合時のオプシンは稀に光活性化したロドプシンと同等の活性を示す**
Apo-opsin exists in equilibrium between a predominantly inactive and a rare highly active state
○佐藤 慎哉 [1,2], Jastrzebska Beata[3], Engel Andreas[3], Palczewski Krzysztof[3,4], Kefalov Vladimir J.[1]（[1]セントルイス・ワシントン大, [2]京都大・院生命科学, [3]ケースウエスタンリザーブ大, [4]カルフォルニア大アーバイン校）
Shinya Sato[1,2], Beata Jastrzebska[3], Andreas Engel[3], Krzysztof Palczewski[3,4], Vladimir J. Kefalov[1] ([1]*DOVS, Washington Univ.*, [2]*Grad. Sch. Biostudies., Kyoto Univ.*, [3]*Case Western Reserve Univ.*, [4]*UC Irvine*)

9:00　**篠田　肇**　3Pos180
1YA0900　**酸性細胞環境内の超解像イメージング応用に向けた耐酸性可逆的光スイッチング緑色蛍光タンパク質の開発**
Acid-tolerant Reversibly Switchable Green Fluorescent Protein for Super-resolution Imaging in Acidic Conditions
○篠田 肇 [1,2], Lu Kai[3], 中島 良介 [3], 和沢 鉄一 [3], 野口 滉介 [2], 松田 知己 [2,3], 永井 健治 [2,3]（[1]理研・開拓研究本部, [2]阪大・工, [3]阪大・産研）
Hajime Shinoda[1,2], Kai Lu[3], Ryosuke Nakashima[3], Tetsuichi Wazawa[3], Kosuke Noguchi[2], Tomoki Matsuda[2,3], Takeharu Nagai[2,3] ([1]*CPR, Riken*, [2]*Grad. Sch. Eng., Osaka Univ.*, [3]*ISIR, Osaka Univ.*)

9:15　武井　洋大　1Pos262

1YA0915 Intron seqFISH enables transcriptome-wide visualization of genome organization and nascent transcription in single cells

Yodai Takei[1], Sheel Shah[2], Wen Zhou[1], Eric Lubeck[3], Jina Yun[1], Chee-Huat Linus Eng[1], Noushin Koulena[1], Christopher Cronin[1], Christoph Karp[1], Eric Liaw[2], Mina Amin[4], Long Cai[1] ([1]*California Institute of Technology*, [2]*University of California, Los Angeles*, [3]*Stanford University*, [4]*University of California, Riverside*)

9:30　Tran Phuoc Duy　3Pos025

1YA0930 フレキシブルドッキングによる結合自由エネルギーと速度定数計算

Calculation of binding free energy and kinetic rates with flexible protein docking

○Tran Duy, 北尾 彰朗（東工大・生命理工）

Duy Tran, Akio Kitao (*Tokodai, Grad. Life Sci. Tech.*)

9:45　堤　研太　3Pos016

1YA0945 クライオ電子顕微鏡による多剤排出ポンプ複合体 MexAB–OprM の構造解析

The wild-type structures of MexAB-OprM multidrug efflux pump revealed by cryo-electron microscopy

○堤 研太 [1], 米原 涼 [1], 池田 悦子 [1], 宮崎 直幸 [1,2], 前田 晋太朗 [1,3], 岩崎 憲治 [1,2], 中川 敦史 [1], 山下 栄樹 [1]（[1]阪大・蛋白研, [2]筑波大・生存ダイナミクス研, [3]スクリプス研究所）

Kenta Tsutsumi[1], Ryo Yonehara[1], Etsuko Ishizaka-Ikeda[1], Naoyuki Miyazaki[1,2], Shintaro Maeda[1,3], Kenji Iwasaki[1,2], Atsushi Nakagawa[1], Eiki Yamashita[1] ([1]*IPR, Univ. Osaka*, [2]*TARA, Univ. Tsukuba*, [3]*The Scripps Research Inst.*)

10:00　中村　彰彦　2Pos132

1YA1000 結晶性キチン加水分解酵素は背水の陣で進むブラウニアンモーターである

Crystalline chitin hydrolase is a Burnt-bridge Brownian motor

○中村 彰彦 [1,2], 岡崎 圭一 [1], 古田 忠臣 [3], 櫻井 実 [3], 飯野 亮太 [1,2]（[1]自然科学研究機構 分子科学研究所, [2]総合研究大学院大学, [3]東京工業大学）

Akihiko Nakamura[1,2], Kei-ichi Okazaki[1], Tadaomi Furuta[3], Minoru Sakurai[3], Ryota Iino[1,2] ([1]*Institute for Molecular Science*, [2]*SOKENDAI*, [3]*Tokyo Institute of Technology*)

10:15　丸山慎太郎　2Pos118

1YA1015 The combination of high-speed atomic force microscopy and X-ray crystallography reveals rotary catalysis of a shaftless V1 motor

Shintaro Maruyama[1], Motonori Imamura[2], Takayuki Uchihashi[3,4], Kazuya Nakamoto[1], Kenji Mizutani[5], Lica Fabiana Imai[1], Kano Suzuki[1], Yoshiko Ishizuka-Katsura[6], Tomomi Someya-Kimura[6], Mikako Shirouzu[1,6,7], Ichiro Yamato[1,7], Toshio Ando[2], Takeshi Murata[1,8] ([1]*Grad. Sch. Sci., Univ. Chiba*, [2]*WPI Nano Life Sci. Inst., Univ. Kanazawa*, [3]*JST, CREST*, [4]*Dep. Phys., Univ. Nagoya*, [5]*Grad. Sch. Med. Life. Sci., Univ. Yokohama*, [6]*DSSB, RIKEN*, [7]*Ind. Sci. Tokyo Univ. Sci.*, [8]*PREST, JST*)

10:30　柳沼　秀幸　2Pos278

1YA1030 定量的 ATP イメージングを用いた細胞の代謝状態の空間的相関の解析

Spatial correlation of metabolic states in mammalian cells revealed by quantitative single-cell ATP imaging

○柳沼 秀幸 [1,2], 岡田 康志 [1,3]（[1]理研・BDR, [2]東大院・工, [3]東大院・理）

Hideyuki Yaginuma[1,2], Yasushi Okada[1,3] ([1]*BDR, Riken*, [2]*Grad. Sch. of Eng., Univ. of Tokyo*, [3]*Grad. Sch. of Sci., Univ. of Tokyo*)

10:45　**山置　佑大**　1Pos091

1YA1045　**In-cell NMR 法を用いたヒト生細胞内核酸の構造およびダイナミクスの評価**

Evaluation of the structure and dynamics of nucleic acids inside the living human cells by in-cell NMR spectroscopy

○山置 佑大[1], 永田 崇[1,2], 清石 彩華[2], 三宅 雅之[2], 加納 ふみ[3], 村田 昌之[3,4], 片平 正人[1,2] ([1]京大・エネルギー理工学研究所, [2]京大・院エネルギー科学, [3]東工大・科学技術創成研究院, [4]東大・院総合文化)

Yudai Yamaoki[1], Takashi Nagata[1,2], Ayaka Kiyoishi[2], Masayuki Miyake[2], Fumi Kano[3], Masayuki Murata[3,4], Masato Katahira[1,2] ([1]*Inst. Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Energy Sci., Kyoto Univ.*, [3]*Inst. Innovative Res., Tokyo Inst. Technol.*, [4]*Grad. Sch. Arts and Sci., Univ. Tokyo*)

カレントトピックス　Current Topics

1 日目（9 月 24 日（火））／Day 1（Sep. 24 Tue.）

13:40〜16:20　A 会場（4F 天蘭）／Room A (4F Tenran)
1CAP　タンパク質の構造・機能・デザイン
Proteins: structure・function・design

オーガナイザー：村上 聡（東工大），山下 敦子（岡山大）
Organizers: Satoshi Murakami (Tokyo Inst Tech), Atsuko Yamashita (Okayama Univ)

1CAP-01　急速に拡大する微生物型ロドプシンワールド
Fast-expanding microbial rhodopsin world
Keiichi Inoue (*Inst. Solid State Phys., Univ. Tokyo*)

1CAP-02　生体光物理の挑戦：静的な孤立系から組織化された動的な集合体の理解へ
Next challenge in biological photophysics: From static/isolated protein to dynamic/assembled protein network
Toru Kondo (*Tohoku University*)

1CAP-03　CRISPR-Cas9 の立体構造と機能改変
Structure and Engineering of CRISPR-Cas9
Hiroshi Nishimasu (*The University of Tokyo*)

1CAP-04　生物分子モーターとその集合体の再デザイン
Re-design of biomolecular motors and their systems
Ken'ya Furuta (*NICT*)

1CAP-05　相分離研究の最近の動向
Current trend of phase separation
Tomoshi Kameda, Yoichi Kurumida, Yusuke Nakamichi, Keisuke Ikeda, Ryo Kitahara, Kiyoto Kamagata (*AIRC, AIST*)

2 日目（9 月 25 日（水））／Day 2（Sep. 25 Wed.）

8:30〜11:10　A 会場（4F 天蘭）／Room A (4F Tenran)
2CAA　細胞の生物物理学
Biophysics of the cells

オーガナイザー：杉村 薫（京大），武井 洋大（カリフォルニア工科大）
Organizers: Kaoru Sugimura (Kyoto Univ), Yodai Takei (Caltech)

2CAA-01　バイオメディカル、細胞生物学、生理学分野におけるナノ温度計測の近況
Current nanothermometry in cell biology and physiology
Madoka Suzuki (*Institute for Protein Research, Osaka University*)

2CAA-02　細胞の中ではたらく物理的な力
Physical forces in the cell
Hirokazu Tanimoto (*Yokohama City University*)

2CAA-03　The mechanics of nature behind the multicellular tissue structure
Kaoru Sugimura (*Institute for Integrated Cell-Material Sciences (WPI-iCeMS), Kyoto Univ.*)

2CAA-04　細胞外体液動態を介した分泌タンパク質の新たな制御機構
Novel extracellular fluid mechanism for regulation of secreted proteins in Xenopus laevis
Hidehiko Inomata (*RIKEN BDR*)

2CAA-05　Recent Advances in Origins of Life Research by Biophysicists
Tony Z. Jia (*Earth-Life Science Institute, Tokyo Institute of Technology and Blue Marble Space Institute of Science*)

14:10〜17:20　A 会場（4F 天蘭）／Room A (4F Tenran)
2CAP　物理理論と実験のクロスオーバー
Crossover of physics theories and experiments

オーガナイザー：柳澤 美穂（東大），田端 和仁（東大）
Organizers: Miho Yanagisawa (Univ Tokyo), Kazuhito Tabata (Univ Tokyo)

2CAP-01　高分子混雑と細胞サイズ閉じ込めの協奏による特異な相挙動
Unique phase behavior in cell size space: Synergistic effect of molecular crowding and confinement
Miho Yanagisawa (*Graduate School of Arts and Sciences, The University of Tokyo*)

2CAP-02　合成生物学は生物実験をどう変えるか　―生物物理学の視点から―
“Dream” experiment in living cells? Synthetic biology may help in near future!
Hideki Nakamura (*The Johns Hopkins University*)

2CAP-03　神経細胞軸索輸送の分子モーターカウンティング
Molecular motor counting for neuronal cargo transport
Kumiko Hayashi[1,2] ([1]*Sch. Eng., Tohoku Univ.*, [2]*PRESTO, JST*)

2CAP-04　多細胞系の非平衡物理モデルと実験
Physical properties of homeostatic and active tissues
Kyogo Kawaguchi (*RIKEN CPR*)

2CAP-05　細胞建築学：物理学と遺伝学の一交差点
Architectonics of the cell, as a crossroad of physics and genetics
Akatsuki Kimura[1,2] ([1]*Cell Arch Lab, National Institute of Genetics*, [2]*Dept Genetics, SOKENDAI*)

2CAP-06　神経ダイナミクスの埋め込みと予測
Embedding and Predicting Neural Dynamics
Taro Toyoizumi (*RIKEN Center for Brain Science*)

3 日目（9 月 26 日（木））／Day 3（Sep. 26 Thu.）

8:30〜11:10　A 会場（4F 天蘭）／Room A (4F Tenran)
3CAA　最先端計測技術
State-of-the-art measurement

オーガナイザー：南後 恵理子（京大・理研），坂内 博子（理研）
Organizers: Eriko Nango (Kyoto Univ & RIKEN), Hiroko Bannai (RIKEN)

3CAA-01　X 線自由電子レーザーによるタンパク質分子動画撮影
Molecular movies of proteins at work by X-ray free electron lasers
Eriko Nango[1,2], So Iwata[1,2] ([1]*Graduate School of Medicine Kyoto University*, [2]*RIKEN SPring-8 Center*)

3CAA-02　クライオ電子顕微鏡法による生物試料の構造解析
Structural Analysis of Biological Samples by Cryogenic Electron Microscopy
Kaoru Mitsuoka (*Research Center for Ultra-High Voltage Electron Microscopy*)

3CAA-03　生細胞の超解像度機能イメージング
Super resolution live cell functional imaging
Yasufumi Takahashi (*Kanazawa university, NanoLSI*)

3CAA-04　機械学習が駆動する次世代ハイコンテンツ解析で何を「見る」か
Machine learning-driven high content analysis "Beyond seeing is believing
Sadao Ota (*Research Center for Advanced Science and Technology*)

3CAA-05 大脳皮質から脳深部までの包括的な脳活動計測が可能な完全埋め込み型フレキシブル・ストレッチャブ集積回路システム

Brain-implanted flexible and stretchable integrated circuit system for comprehensively monitoring brain activities from cerebral cortex to deep brain regions

Tsuyoshi Sekitani (*The Institute of Scientific and Industrial Research, Osaka University*)

8:30〜11:10　B 会場（4F 天玉）／Room B (4F Tengyoku)

1SBA　ゲノム機能発現の統合的理解に向けた多角的アプローチ
Integrative approaches towards understanding of gene expression

オーガナイザー：森 貴治（理化学研究所），関根 俊一（理化学研究所）
Organizers: Takaharu Mori (RIKEN), Shun-ichi Sekine (RIKEN)

Recent advances in experimental and theoretical techniques have enabled us to understand detailed mechanisms of gene expression. Since conventional approaches in vitro are usually not enough to fully understand them, integrative approaches combining various techniques are essential to reveal dynamic structures of proteins and DNA in the cell at the molecular level. In this symposium, we discuss how hybrid approaches using the X-ray crystallography, cryo-electron microscopy, single-molecule imaging, and molecular simulations can contribute to the integrative understandings of genome functions.

1SBA-1　Chromatin dynamics and transcription
Kazuhiro Maeshima, Kayo Hibino, Ryosuke Nagashima (*Genome Dynamics Laboratory, National Institute of Genetics*)

1SBA-2　真核生物における転写メディエーター複合体の構造機能解析
Structural and functional basis of the Mediator complex in the eukaryotic transcriptional system
○野澤 佳世（東京大学・定量生命科学研究所）
Kayo Nozawa (*The University of Tokyo, Institute for Quantitative Biosciences*)

1SBA-3　RNA ポリメラーゼ II によるヌクレオソーム転写のメカニズム
Structural basis of nucleosome transcription by RNA polymerase II
○関根 俊一（理研 BDR）
Shun-ichi Sekine (*RIKEN BDR*)

1SBA-4　クライオ電顕と MD 計算による RNA ポリメラーゼ II 伸長状態複合体の動態解析
Dynamic structures of the RNA polymerase II elongation complex by cryo-EM and MD approaches
○森 貴治 [1], 江原 晴彦 [2], 関根 俊一 [2], 杉田 有治 [1,3,4]（[1]理研 杉田理論分子科学, [2]理研 BDR (横浜), [3]理研 BDR (神戸), [4]理研 R-CCS）
Takaharu Mori[1], Haruhiko Ehara[2], Shun-ichi Sekine[2], Yuji Sugita[1,3,4] ([1]*RIKEN Theor. Mol. Sci. Lab.*, [2]*RIKEN BDR (Yokohama)*, [3]*RIKEN BDR (Kobe)*, [4]*RIKEN R-CCS*)

1SBA-5　Allostery of Nucleosomal DNA for Transcription Factor Binding
Cheng Tan, Shoji Takada (*Kyoto University*)

1SBA-6　(1Pos082) 大腸菌非六量体型 DNA ヘリカーゼ UvrD C 末端欠損変異体の 1 分子イメージング
(1Pos082) Single-molecule imaging of a non-hexameric *Escherichia coli* helicase UvrD mutant lacking C-terminal residues
○横田 浩章（光産創大・光バイオ）
Hiroaki Yokota (*Biophotonics Lab., Grad. Sch. Creation New Photon. Indust.*)

8:30〜11:10　C 会場（4F 天樹）／Room C (4F Tenjyu)

1SCA　階層を超えた柔軟な場と空間の活用：生命システムが持つ可能性を探る
Utilization of soft compartments/interfaces from nano to macroscale: Exploring the potential of living systems

オーガナイザー：佐藤 佑介（東京工業大学），森田 雅宗（産業総合技術研究所），鈴木 勇輝（東北大学）
Organizers: Yusuke Sato (Tokyo Institute of Technology), Masamune Morita (AIST), Yuki Suzuki (Tohoku University)

Fundamental reactions of life occur mainly in a space surrounded by a biological membrane or on its interface. Unraveling phenomena that are specific to such a 'soft' compartment/interface not only leads to understanding the living systems but also gives us clues in designing artificial biosystems more rationally. In this symposium, we will discuss the potential and perspectives of the use of soft compartments/interfaces at several hierarchical levels, from nano to macroscale.

1SCA-1　DNA origami lattices self-assembled on lipid bilayer membranes
Yuki Suzuki (*FRIS, Tohoku Univ.*)

1SCA-2　(1Pos074) 脂質分子の混み合い効果による膜貫通タンパク質結晶化の検討
(1Pos074) Crystallization of transmembrane protein driven by molecular crowding effect of lipids: Theoretical estimation by using a simple model
○須田 慶樹 [1], 末松 安由美 [2], 秋山 良 [1]（[1]九州大学理学府, [2]九州産業大学）
Keiju Suda[1], Ayumi Suematsu[2], Ryo Akiyama[1] ([1]*Kyushu University, Sci.*, [2]*Kyushu Sangyo University, Science and Engineering*)

1SCA-3　生理学的等温条件下における細胞サイズリポソーム内での特定配列を持つ DNA 分子の増幅
Amplification of specific DNA molecules inside giant unilamellar vesicles at isothermal and physiological temperature
○佐藤 佑介 [1], 小宮 健 [1], 川又 生吹 [2], 村田 智 [2], 野村 M. 慎一郎 [2]（[1]東工大・情報理工, [2]東北大・院工）
Yusuke Sato[1], Ken Komiya[1], Ibuki Kawamata[2], Satochi Murata[2], Shin-ichiro M. Nomura[2] ([1]*Sch. Comput. Tokyo Tech*, [2]*Grad. Sch. Eng., Tohoku Univ.*)

1SCA-4　(1Pos078) cDNA ディスプレイとセルソーターの利用による新規リポソームポア形成ペプチドの創製
(1Pos078) Novel pore-forming peptides assembling in liposome membranes selected by combining cDNA display method with cell sorter system
○根本 直人 [1], 宮嶋 俊樹 [1], 吉延 武留 [1], 關谷 悠介 [2], 川野 竜司 [2] ([1]埼玉大学大学院 理工研, [2]東京農工大 生命工学）
Naoto Nemoto[1], Toshiki Miyajima[1], Takeru Yoshinobu[1], Yusuke Sekiya[2], Ryuji Kawano[2] ([1]*Grad. Sci. Eng., Saitama Univ.*, [2]*Dept. Biotech. Life. Sci., Tokyo Univ. Agr. Tech*)

1SCA-5　(1Pos287) 光からエネルギーを合成しタンパク質合成をする人工光合成細胞の構築
(1Pos287) Artificial photosynthetic cell producing energy for protein synthesis
ベルハヌ サミュエル [2], 上田 卓也 [3], ○車 兪澈 [1]（[1]海洋研究開発機構, [2]東工大・地球生命研究所, [3]東大院・新領域）
Samuel Berhanu[2], Takuya Ueda[3], **Yutetsu Kuruma**[1] ([1]*JAMSTEC*, [2]*ELSI, Titech*, [3]*Grad. Sch. of Front. Sci., Univ. of Tokyo*)

1SCA-6　ベシクル型細胞モデルにおけるこみあい効果
A study of crowding effect in a cell model using a statistical mechanics approach
○夏目 ゆうの [1,2]（[1]日女大・理, [2]生命創成セ）
Yuno Natsume[1,2] ([1]*Fac. Sci., Japan Women's Univ.*, [2]*ExCELLS*)

1SCA-7　Construction of cell-containing synthetic vesicles for bottom-up synthetic biology
Masamune Morita (*Biomed. Res. Inst. (BMRI), AIST*)

1SCA-8　計算折り紙による 3 次元細胞立体構造
3D Cell Structure Optimized by Computational Origami
○繁富 香織（北海道大学）
Kaori Kuribayashi-Shigetomi (*Hokkaido University*)

8:30〜11:10　D 会場（4F 天葉）／Room D (4F Tenyo)
1SDA　クライオ電子顕微鏡でできること、できないこと　−構造生命科学の最先端−
What can or cannot do by cryo-EM? The forefront of Structural Life Science

オーガナイザー：吉川 雅英（東京大学），中川 敦史（大阪大学）
Organizers: Masahide Kikkawa (The University of Tokyo), Atsushi Nakagawa (Osaka University)

“Resolution revolution” of cryo-electron microscopy (cryo-EM) dramatically proceeded the field of structural life science. Cryo-EM is one of the best tools for structure determination of biological macromolecules, however, it is not a perfect tool to understand living system at atomic resolution. We are organizing this symposium to discuss direction of structural life science using combination of various advanced techniques including cutting-edge cryo-EM.

はじめに
Opening Remarks
中川 敦史（大阪大）
Atsushi Nakagawa (*Osaka Univ.*)

1SDA-1　Structural transition of nucleosome during RNA polymerase II transcription revealed by cryo-EM
Tomoya Kujirai[1,2], Haruhiko Ehara[2], Mikako Shirouzu[2], Shun-ichi Sekine[2], Hitoshi Kurumizaka[1,2] ([1]*IQB, Univ. of Tokyo*, [2]*RIKEN BDR*)

1SDA-2　タンパク質の柔軟な構造を高速原子間力顕微鏡で可視化する
Visualizing flexibility in protein structures by high-speed atomic force microscopy
○柴田 幹大 [1,2]（[1]金沢大・WPI-NanoLSI, [2]金沢大・新学術創成）
Mikihiro Shibata[1,2] ([1]*WPI-NanoLSI, Kanazawa Univ.*, [2]*InFiniti, Kanazawa Univ.*)

1SDA-3　クライオ電子顕微鏡解析によって明らかになったミトコンドリア膜透過装置の構造と機能
Near-atomic resolution structure of the mitochondrial protein import gate
○荒磯 裕平 [1], 包 明久 [2], 今井 賢一郎 [3], 阪上 春花 [4], 塩田 拓也 [5], 柚木 芳 [4], 鈴木 純子 [4], 河野 慎 [4], 吉川 雅英 [2], 遠藤 斗志也 [4]（[1]金沢大・保健, [2]東大・医, [3]産総研, [4]京産大・生命科, [5]宮崎大）
Yuhei Araiso[1], Akihisa Tsutsumi[2], Kenichiro Imai[3], Haruka Sakaue[4], Takuya Shiota[5], Kaori Yunoki[4], Junko Suzuki[4], Shin Kawano[4], Masahide Kikkawa[2], Toshiya Endo[4] ([1]*Grad. Sch. of Med. Sci., Kanazawa Univ.*, [2]*Grad. Sch. of Med., Univ. of Tokyo*, [3]*AIST*, [4]*Fac. of Life Sci., Kyoto Sangyo Univ.*, [5]*OPTT, Univ. of Miyazaki*)

1SDA-4　Microsystem for single molecule analysis of membrane proteins
Rikiya Watanabe (*CPR, RIKEN*)

1SDA-5 Cryo-EM structures of photosystem II-antenna supercomplexes
Fusamichi Akita[1,2], Ryo Nagao[1], Koji Kato[1], Naoyuki Miyazaki[3], Jian-Ren Shen[1] ([1]*RIIS, Okayama Univ.*, [2]*PRESTO, JST*, [3]*TARA, Univ. Tsukuba*)

1SDA-6 光合成型複合体 I がフェレドキシン依存性を示す構造基盤
Structural Basis for the Ferredoxin-dependency of Photosynthetic Complex I
○栗栖 源嗣 [1], 田中 秀明 [1], シューラー ヤン [2], 小沼 剛 [3], 池上 貴久 [3], ノバクチック マーク [4]（[1]阪大蛋白研, [2]マックスプランク生化学研究所, [3]横市大・院生命医科学, [4]ルール大学ボーフム）
Genji Kurisu[1], Hideaki Tanaka[1], Jan M. Schuller[2], Tsuyoshi Konuma[3], Takahisa Ikegami[3], Marc M. Nowaczyk[4] ([1]*Inst. Prot. Res., Osaka Univ.*, [2]*Max Planck Institute of Biochemistry*, [3]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*, [4]*Ruhr University Bochum*)

おわりに
Closing Remarks
中川 敦史（大阪大）
Atsushi Nakagawa (*Osaka Univ.*)

8:30〜11:10　E 会場（4F クリスタルルーム）／Room E (4F Crystal Room)
1SEA　共催：新学術領域研究「遺伝子制御の基盤となるクロマチンポテンシャル」
遺伝子制御の原理に迫るクロマチン動態の物理学
Physics of chromatin dynamics – towards understanding the regulation of gene expression

オーガナイザー：伊藤 由馬（東京工業大学），木村 暁（国立遺伝学研究所）
Organizers: Yuma Ito (Tokyo Institute of Technology), Akatsuki Kimura (National Institute of Genetics)

The life system is maintained by dynamic tuning of metabolisms. Rewiring of the metabolic networks in bacteria, plants or human diseases is considered to be the results of the adaptation of their whole-body metabolisms to environment. The molecular mechanism underlying the metabolic adaptation can be only understood through measuring and analyzing "trans-omic" network, consisting of interactions among molecules across multi-omic layers, such as genome, transcriptome, proteome, and metabolome. Here we hold this symposium to shed light on strategies and obstacles in integrating multiple omic layers to establish trans-omic approaches, and to have discussions with cutting edge researchers in omics research fields.

1SEA-1 1 分子超解像局在顕微鏡法による転写装置とクロマチン構造の相互作用解析
A single-molecule localization approach to quantify the interaction between transcriptional machinery and chromatin structure
○伊藤 由馬, 徳永 万喜洋（東工大・生命理工学院）
Yuma Ito, Makio Tokunaga (*Sch. Life Sci. Tech., Tokyo Tech*)

1SEA-2 単一ヌクレオソームイメージングで迫る分裂期染色体の構築原理
Single nucleosome imaging reveals the physical aspect of the mitotic chromosome condensation
○日比野 佳代 [1,2], 前島 一博 [1,2], 境 祐二 [3]（[1]遺伝研, [2]総研大, [3]東大・医）
Kayo Hibino[1,2], Kazuhiro Maeshima[1,2], Yuji Sakai[3] ([1]*National Institute of Genetics*, [2]*SOKENDAI*, [3]*Grad. Sch. Med., Univ. Tokyo*)

1SEA-3 (1Pos095) オリゴペプチドのアミノ酸配列は DNA compaction と転写活性に著しい違いを引き起こす
(1Pos095) Marked Difference in DNA Compaction and Transcription is Caused by Amino Acid Sequence of Oligopeptide
○秋田谷 龍男 [1], 平松 裕之 [2], 山口 秀明 [3], 久保 康児 [4], 村田 静昭 [4], 神戸 俊夫 [5], 櫨本 紀夫 [6], 吉川 研一 [7], Zinchenko Anatoly[4]（[1]旭川医大・医, [2]名城大・薬, [3]名城大・農, [4]名大・院環境, [5]名大・院医, [6]名市大・院薬, [7]同志社大・生命医）
Tatsuo Akitaya[1], Hiroyuki Hiramatsu[2], Hideaki Yamaguchi[3], Koji Kubo[4], Shizuaki Murata[4], Toshio Kanbe[5], Norio Hazemoto[6], Kenichi Yoshikawa[7], Anatoly Zinchenko[4] ([1]*Asahikawa Med. Univ.*, [2]*Fac. Pharm., Meijo Univ.*, [3]*Fac. Agr. Sci., Meijo Univ.*, [4]*Grad. Sch. Env. Std., Nagoya Univ.*, [5]*Grad. Sch. Med., Nagoya Univ.*, [6]*Grad. Sch. Pharm. Sci., Nggoya City Univ.*, [7]*Fac. Bio. Med. Sci., Doshisah Univ.*)

1SEA-4 クロマチンループを形成しないヌクレオソーム排他 DNA 配列によるインスレーター活性
Insulator Activities of Nucleosome-Excluding DNA Sequences Without Chromatin Loop Formations
○粟津 暁紀 [1], 松島 佑樹 [2], 坂本 尚昭 [1]（[1]広島大学大学院統合生命科学研究科, [2]広島大学大学院統合生命科学研究科）
Akinori Awazu[1], Yuki Matsushima[2], Naoaki Sakamoto[1] ([1]*Dept. of Math. and Life Sciences, Hiroshima Univ.*, [2]*Dept. of Math. and Life Sciences, Hiroshima Univ.*)

1SEA-5 (1Pos239) Molecular Dynamics of Nucleosome Assembly
Giovanni Brandani, Shoji Takada, Cheng Tan (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

1SEA-6 (1Pos088) エピジェネティック修飾をもつクロマチンのモデルにおける不連続相転移
(1Pos088) Discontinuous Phase Transition in a Chromatin Model with Epigenetic Modification
○足立 景亮, 川口 喬吾（理研 BDR）
Kyosuke Adachi, Kyogo Kawaguchi (*RIKEN BDR*)

1SEA-7 Transcription dynamics of DNA at interfaces
Tetsuya Yamamoto (*Nagoya Univ., Dep. of Mat. Phys.*)

1SEA-8 クロマチンの高次構造とダイナミクス ～高分子物理の視点から
Structure and dynamics of chromatins: perspective from polymer physics
○坂上 貴洋 [1,2]（[1]青山学院大学 理工学部 物理・数理学科, [2]JST さきがけ）
Takahiro Sakaue[1,2] ([1]*Department of Physics and Mathematics, Aoyama Gakuin University*, [2]*JST, PRESTO*)

8:30〜11:10　F 会場（4F マーブルルーム）／Room F (4F Marble Room)

1SFA　共催：新学術領域研究「共鳴誘導で革新するバイオイメージング」
生体機能の「ありのまま」の可視化と理解へ
〜共鳴する生命現象と光技術〜
Toward "Ari-No-Mama" visualization to reveal biological functions -Resonance between life science and optical technology~

オーガナイザー：宮脇 敦史（理化学研究所），根本 知己（北海道大学）
Organizers: Atsushi Miyawaki (RIKEN), Tomomi Nemoto (Hookaido University)

Visualization and quantitative analysis of in-vivo events are important for the understanding of the molecular basis and emergence of biological functions. Moreover, recent developments in optical technology such as nonlinear optics and lasers as well as new probes have led to the development of new bioimaging in life sciences. In this symposium, we will discuss young and energetic researchers about the latest achievements and future prospects about new methods developed from the resonance between life sciences and optical sciences.

1SFA-1　Genetically encoded tools for brain sciences
Atsushi Miyawaki (*RIKEN*)

1SFA-2　**蛍光全脳イメージングのための連続断層イメージング法 FAST**
Block-face serial microscopy tomography for whole-brain fluorescence imaging
○勢力 馨 [1,2], 橋本 均 [1,3,4,5]（[1]大阪大・薬・神経薬理, [2]大阪大・国際共創大学院, [3]大阪大・子どものこころ, [4]大阪大・データビリティフロンティア機構, [5]大阪大・先導的学際研究機構）
Kaoru Seiriki[1,2], Hitoshi Hashimoto[1,3,4,5] ([1]*Lab. Mol. Neuropharmacol., Grad. Sch. Pharmaceutical Sci., Osaka Univ.*, [2]*Interdisciplinary Program for Biomedical Sci., Inst. Transdisciplinary Graduate Degree Programs, Osaka Univ.*, [3]*Mol. Res. Cent. Children's Mental Development, United Grad. Sch. Child Development, Osaka Univ.*, [4]*Div. Biosci., Inst. Datability Sci., Osaka Univ.*, [5]*Transdimensional Life Imaging Div., Inst. Open and Transdisciplinary Res. Initiatives, Osaka Univ.*)

1SFA-3　**非回折と自己湾曲特性を用いた光ニードル顕微鏡における 3 次元イメージング**
Three-dimensional imaging in light needle microscopy utilizing non-diffraction and self-bending characteristics
○小澤 祐市, 佐藤 俊一（東北大多元所）
Yuichi Kozawa, Shunichi Sato (*IMRAM, Tohoku UNiv.*)

1SFA-4　**広視野 2 光子デジタル走査ライトシート顕微鏡とメダカ胚全身イメージングへの応用**
Wide-field 2-photon light-sheet microscopy and its application to whole body imaging of medaka embryos
○齋藤 卓, 今村 健志（愛媛大学）
Takashi Saitou, Takeshi Imamura (*Ehime University*)

1SFA-5 線虫の全脳機能的イメージングのための要素技術と全神経活動の解析
Bio-image informatics for whole brain activity imaging and analysis of neural activity of C. elegans
○豊島 有[1], Wu Stephen[3], 金森 真奈美[1], 佐藤 博文[1], 張 文瑄[1], 村上 悠子[2], 大江 紗[2], 德永 旭将[4], 広瀬 修[5], 久下 小百合[2], 寺本 孝行[2], 岩崎 唯史[6], 吉田 亮[3], 石原 健[2], 飯野 雄一[1]（[1]東大・院理・生物科学, [2]九大・院理・生物科学, [3]統計数理研究所, [4]九工大・大学院情報工学研究院, [5]金沢大・生命理工学系, [6]茨城大・工・知能システム）
Yu Toyoshima[1], Stephen Wu[3], Manami Kanamori[1], Hirofumi Sato[1], Moon Sun Jang[1], Yuko Murakami[2], Suzu Oe[2], Terumasa Tokunaga[4], Osamu Hirose[5], Sayuri Kuge[2], Takayuki Teramoto[2], Yuishi Iwasaki[6], Ryo Yoshida[3], Takeshi Ishihara[2], Yuichi Iino[1] ([1]*Dept of Biological Sciences, Grad Sch of Science, Univ of Tokyo*, [2]*Dept of Biology, Facl of Sciences, Kyushu Univ*, [3]*Inst of Statistical Mathematics, Research Organization of Information and Systems*, [4]*Dept of Systems Design and Informatics, Facl of Computer Science and Systems Engineering, Kyushu Inst of Technology*, [5]*Facl of Electrical and Computer Engineering, Inst of Science and Engineering, Kanazawa Univ*, [6]*Dept. of Mec. Eng., Grad. Sch. of Sci. and Eng., Ibaraki Univ.*)

1SFA-6 Bilateral Domain 画像処理
Bilateral Domain Image Processing
○吉澤 信（理化学研究所 光量子工学研究センター 画像情報処理研究チーム）
Shin Yoshizawa (*IPRT, RAP, RIKEN*)

8:30～11:10　G 会場（4F アイボリールーム）／Room G (4F Ivory Room)
1SGA　筋・血管系のマルチスケールメカノバイオロジーの最前線
Frontiers in multi-scale mechanobiology of muscle and vascular system

オーガナイザー：岩城 光宏（理化学研究所），原 雄二（京都大学）
Organizers: Mitsuhiro Iwaki (RIKEN), Yuji Hara (Kyoto University)

The field of mechanobiology has grown dramatically in the past decade and diverse biological systems are currently targeted. Especially, importance of physical stimuli and the response in muscle and vascular system is well known at the phenomenological level, however, the molecular mechanism and multiscale relationships between molecules and cells, tissues or organ is still elusive. This symposium will provide an overview of the latest findings in the field, revealing the relationship between physical stimuli and activities of muscle and vascular system at each hierarchy.

1SGA-1 Thick filament activation through a molecular-based mechanosensing, regulates forces in mathematical models of trabecula and ventricle
Lorenzo Marcucci[1,3], Takumi Washio[2], Toshio Yanagida[3] ([1]*Department of Biomedical Sciences, Padova University, Italy*, [2]*Graduate School of Frontier Sciences, The University of Tokyo, Japan*, [3]*Center for Biosystems Dynamics Research, RIKEN, Japan*)

1SGA-2 (1Pos108) 心筋細胞に備わる収縮リズム恒常性の分子機構の解明
(1Pos108) Elucidation of molecular mechanism of contraction rhythm homeostasis in cardiac myocytes
○新谷 正嶺[1], 鷲尾 巧[2]（[1]中部大 生命健康科学部 生命医科学科, [2]東京大学 新領域創成科学研究科）
Seine Shintani[1], Takumi Washio[2] ([1]*Department of Biomedical Sciences, College of Life and Health Sciences, Chubu University*, [2]*Graduate School of Frontier Sciences, the University of Tokyo*)

1SGA-3 DNA ナノデバイスと高解像 1 分子イメージング技術を活用した心臓のメカノバイオロジー
Mechanobiology of Heart Revealed with DNA Nano-device and Nanometer-precision Single-molecule Imaging
○岩城 光宏 [1,2]（[1]理研・生命機能科学センター, [2]阪大・院生命機能）
Mitsuhiro Iwaki[1,2] ([1]*RIKEN, BDR*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

1SGA-4 Myosin filament regulation by mechanosensing in skeletal and cardiac muscle
Vincenzo Lombardi (*PhysioLab*)

1SGA-5 骨格筋再生における機械受容イオンチャネル PIEZO1 の役割
Role of a mechanosensitive cation channel PIEZO1 in skeletal muscle regeneration
○原 雄二 [1,2], 平野 航太郎 [1], 高林 征史 [1], 土谷 正樹 [1], 梅田 眞郷 [1]（[1]京都大学大学院工学研究科 合成・生物化学専攻 生体認識化学分野, [2]AMED PRIME）
Yuji Hara[1,2], Kotaro Hirano[1], Seiji Takabayashi[1], Masaki Tsuchiya[1], Masato Umeda[1] ([1]*Graduate School of Engineering, Kyoto University*, [2]*AMED PRIME*)

1SGA-6 Mechano-protective roles of sugar chain in skeletal muscle
Motoi Kanagawa (*Kobe Univ. Grad. Sch. Med.*)

1SGA-7 Lipid bilayer membrane mediated mechanotransduction in vascular endothelial cells
Kimiko Yamamoto[1], Joji Ando[2] ([1]*System Physiology, Graduate School of Medicine, The University of Tokyo*, [2]*Laboratory of Biomedical Engineering, School of Medicine, Dokkyo Medical University*)

1SGA-8 (1Pos268) 細胞内動態をサブセルレベルで制御する温和な NanoHeating 技術
(1Pos268) A Thermodynamic Tool for Mechanobiology Research: Mild Nanoheating Technology to Alter Subcellular Dynamics
○新井 敏 [1], ファーディ ナンデス [2]（[1]早大・理工研, [2]早稲田シンガポール研）
Satoshi Arai[1], Nandus Ferdi[2] ([1]*Res. Inst. Sci. Eng., Waseda Univ.*, [2]*WABIOS*)

8:30〜11:10　H 会場（4F アンバールーム）／Room H (4F Amber Room)

1SHA　静水圧刺激により生命機能を操作する
Control of biological functions with hydrostatic pressure stimulation

オーガナイザー：畑 宏明（東京工業大学），西山 雅祥（近畿大学）
Organizers: Hiroaki Hata (Tokyo Institute of Technology), Masayoshi Nishiyama (Kindai University)

Living organisms change the shape and activity as a response of external forces. The response to the force can be found in biomolecules composing life organisms. Hydrostatic pressure has been used as a tool to apply isotropic forces for investigating the force response of molecular structures and functions. However, pressures used in the previous studies were often high where the biomolecules denature. Recent studies show that much lower pressures keeping structures of biomolecules can affect cellular functions. In this symposium, we will discuss about mechanical control of cellular functions by pressure and the mechanism underlying the pressure effect.

1SHA-1 高圧力による生きた細胞内の分子機械の活性化
Activation of molecular machinery in living cells using high-pressure techniques
○西山 雅祥（近大・理工）
Masayoshi Nishiyama (*Kindai Univ.*)

1SHA-2 (1Pos147) RhoA activation induces cell cycle exit and differentiation of skin cancer cells
Oleg Dobrokhotov, Masahiro Sokabe, Hiroaki Hirata (*Nagoya Univ., Grad. Sch. Med.*)

1SHA-3 (1Pos146) Direct observation of cell mechanics under high hydrostatic pressure
Masatoshi Morimatsu, Keiji Naruse (*Grad. Sch. of Med., Dent. and Pharma. Sci., Okayama Univ.*)

1SHA-4 (1Pos253) 高圧力下で早くなるシアノバクテリアの概日周期
(1Pos253) Pressure accelerates the circadian clock of cyanobacteria
○北原 亮[1], 大山 克明[2], 川村 宇宙[2], 三橋 景汰[2], 北沢 創一郎[1], 安永 和寛[1], 相良 夏乃[1], 藤本 恵[2], 寺内 一姫[2]（[1]立命館大・薬, [2]立命館大・生命）
Ryo Kitahara[1], Katsuaki Oyama[2], Takahiro Kawamura[2], Keita Mitsuhashi[2], Soichiro Kitazawa[1], Kazuhiro Yasunaga[1], Natsuno Sagara[1], Megumi Fujimoto[2], Kazuki Terauchi[2] ([1]*Pharm. Sci., Ritsumeikan Univ.*, [2]*Life Sci., Ritsumeikan Univ.*)

1SHA-5 細菌べん毛モーター回転に及ぼす高静水圧の影響
Effects of high hydrostatic pressure on the rotation of the bacterial flagellar motor
○川岸 郁朗[1,2]（[1]法政大・生命・生命機能, [2]法政大・ナノテクセンター）
Ikuro Kawagishi[1,2] ([1]*Dept. Frontier Biosci., Hosei Univ.*, [2]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

1SHA-6 圧力感受性変異 YFP の圧力応答の構造基盤
Structural basis of pressure response of a pressure sensitive YFP variant protein
○今田 勝巳[1], 辻井 美香[1], 永江 峰幸[2], 畑 宏明[3], 渡邉 朋信[4], 西山 雅祥[5], 北尾 彰朗[3], 渡邉 信久[2]（[1]阪大・院・理, [2]名大・院・工, [3]東工大・生命, [4]理研・BDR, [5]近大・理工）
Katsumi Imada[1], Mika Tsujii[1], Takayuki Nagae[2], Hiroaki Hata[3], Tomonobu Watanabe[4], Masayoshi Nishiyama[5], Akio Kitao[3], Nobuhisa Watanabe[2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Eng., Nagoya Univ.*, [3]*Sch. LifeSci and Tech,. Tokyo Inst. Tech.*, [4]*BDR, Riken*, [5]*Sch.Sci. and Eng., Kindai Univ.*)

1SHA-7 Pressure effects on protein-protein interactions studied by molecular dynamics simulations
Hiroaki Hata[1], Yasutaka Nishihara[2], Masayoshi Nishiyama[3], Ikuro Kawagishi[4], Akio Kitao[1] ([1]*Dept. of Life Sci. and Tech., Tokyo Tech*, [2]*IMCB, UTokyo*, [3]*Grad Sch. of Sci. and Eng., Kindai Univ.*, [4]*Dept. of Frontier Biosci., Hosei Univ.*)

13:40〜16:20　B 会場（4F 天玉）／Room B (4F Tengyoku)
1SBP　オーストラリアー日本 交流シンポジウム：
生体分子相互作用と介在する力にフォーカスした生物物理学の挑戦
ASB-BSJ Joint Symposium: Current challenges in biophysics centering on biomolecular interactions and the underlying forces

オーガナイザー：西坂 崇之（学習院大学），Marc Kvansakul（La Trobe University）
Organizers: Takayuki Nishizaka (Gakushuin University), Marc Kvansakul (La Trobe University)

To promote greater engagement between the Australian Society for Biophysics and Biophysical Society of Japan we created this exciting symposium featuring presentations spanning the full breath of current challenges in biology that center on biomolecular interactions and the underlying forces driving them. Topics include determination of the forces and interactions between biomolecules including proteins, lipids as well as engineered biomolecular structures to understand the fundamental forces required to maintain or destroy life. Speakers from both societies will discuss advances in understanding the effect of direct attachment on environmental surfaces on the motility and survival of bacteria; principles governing the generation of rhythmic force in animal muscles and cardiac tissues which power life; and lastly, insights into the ability of proteins to change the architecture of lipid bilayers in order to control cellular membranes or contribute to immunity. These exciting presentations provide novel insights into the functions of proteins and their molecular mechanisms of action that control the biological processes that underpin life as we know it.

はじめに
Opening Remarks
西坂 崇之（学習院大）
Takayuki Nishizaka (*Gakushuin Univ.*)

1SBP-1 Microscopic measurements of force and taxis in bacteria/archaea
Takayuki Nishizaka, Daisuke Nakane (*Dept. Phys., Gakushuin Univ.*)

1SBP-2 Biophysical models of physical rupturing of bacterial cells by nano-structured surfaces
Elena Ivanova (*RMIT University*)

1SBP-3 心筋ナノイメージング
Cardiac nano-imaging: from cells to the heart
○福田 紀男（東京慈恵会医科大学・細胞生理学講座）
Norio Fukuda (*Department of Cell Physiology, The Jikei University School of Medicine*)

1SBP-4 How Japanese researchers can get access failing and donor tissue from the Sydney Heart Bank. A viable alternative to using animal models
Cristobal G. dos Remedios, Amy Li, Sean Lal (*Bosch Institute, Discipline of Anatomy & Histology, University of Sydney*)

1SBP-5 リン脂質 flippase による細胞膜変形と細胞機能
Plasma membrane deformation by phospholipid flippase and cellular functions
○申 惠媛（京大・院・薬）
Hye-Won Shin (*Grad Sch Pharm Sci, Kyoto Univ*)

1SBP-6 Structural definition of phospholipid-mediated oligomerization of defensins in fungal and tumour cell lysis
Marc Kvansakul, Mark Hulett, Sofia Caria, Ivan Poon, Michael Jarva, Kha Tran Phan, Fung Lay, Amy Baxter (*La Trobe University*)

おわりに
Closing Remarks
クヴァンサカル マーク（ラ・トローブ大学）
Marc Kvansakul (*La Trobe Univ.*)

13:40〜16:20　C 会場（4F 天樹）／Room C (4F Tenjyu)
1SCP　生物物理で見る脳神経回路
Cutting-edge brain research from a biophysical perspective

オーガナイザー：冨永 貴志（徳島文理大学），Bernd Kuhn（沖縄科学技術大学院大学）
Organizers: Takashi Tominaga (Tokushima Bunri University), Bernd Kuhn (OIST)

The brain is an amazingly complicated and sophisticated information processing device consisting of billions of neurons and thousands of billions of connections in the mammalian brain. To understand how the brain processes information, it is essential to read out or manipulate neuronal activity on all different temporal and spatial scales. In this symposium, various state-of-the-art biophysical methods, especially optical methods, will be presented contributing towards this goal.

1SCP-1 Simultaneous spatio-temporal dendritic voltage/calcium mapping and somatic recording from Purkinje neurons in awake mice
Bernd Kuhn, Christopher J. Roome (*OIST Graduate University*)

1SCP-2 Novel "*in vivo*" two-photon microscopy for vast and longtime neural activity
Tomomi Nemoto (*RIES, Hokkaido Univ.*)

1SCP-3 機能的光干渉断層法とフーリエイメージングによる脳機能構造の 3 次元マイクロ計測
Functional optical coherence tomography with Fourier imaging reveals three-dimensional and micro-scale brain functional structure
○中道 友, 谷藤 学（理研 CBS）
Yu Nakamichi, Manabu Tanifuji (*RIKEN CBS*)

1SCP-4 偏光で解き明かす生細胞内分子アセンブリーのナノ構造とそのダイナミクス
Dissecting nano-scale architectures and dynamics of molecular assemblies in living cells with polarized light
○谷 知己（ウッズホール海洋生物学研究所）
Tomomi Tani (*Marine Biological Laboratory, Woods Hole*)

1SCP-5 (1Pos005) The role of C-terminal carboxylation in α-conotoxin LsIA interactions with human α7 nicotinic acetylcholine receptor *in silico*
Jierong Wen, Andrew Hung (*Sch. Sci., RMIT Univ.*)

1SCP-6 (1Pos266) グルタミン酸受容体を介した植物の長距離 Ca^{2+}シグナル
(1Pos266) Long-distance Ca^{2+} transmission via glutamate receptor channels in plants
○豊田 正嗣 [1,2]（[1]埼玉大・院・理工, [2]University of Wisconsin-Madison）
Masatsugu Toyota[1,2] ([1]*Dept Biochem and Mol Biol, Saitama Univ*, [2]*University of Wisconsin-Madison*)

1SCP-7 光信号で「見る」神経回路のはたらき–膜電位感受性色素（VSD）を中心に
Optical view of the brain neural circuit activity: Voltage-sensitive-dye (VSD) imaging
○冨永 貴志, 冨永 洋子（徳島文理大学神経科学研究所）
Takashi Tominaga, Yoko Tominaga (*Inst. Neurosci., Tokushima Bunri Univ.*)

13:40〜16:20　D 会場（4F 天葉）／Room D (4F Tenyo)
1SDP　蛋白質の溶液物性計測の現状と課題
Current status and issues of protein solution biophysics

オーガナイザー：内山 進（大阪大学），谷中 冴子 （分子科学研究所）
Organizers: Susumu Uchiyama (Osaka University), Saeko Yanaka (Institute for Molecular Science)

The development of biophysico-chemical methods has enabled us the quantitative and systematic analysis of the behavior of biomolecules including their interactions and conformations. Due to the highly sensitive and accurate measurements, even the measurement under in situ conditions are possible. In this symposium, we will introduce solution state measurement techniques, such as analytical ultracentrifugation, thermodynamic measurements, nuclear magnetic resonance. Here we will discuss the recent applications, and future possibility of these methods.

はじめに
Opening Remarks

1SDP-1 溶液中での蛋白質間相互作用の定量的解析法
Quantitative assessments of intermolecular protein mediated interactions in solution
○内山 進 [1,2]（[1]大阪大学大学院工学研究科先端生命工学専攻, [2]自然科学研究機構生命創成探究センター）
Susumu Uchiyama[1,2] ([1]*Department of Biotechnology, Graduate School of Engineering, Osaka University*, [2]*ExCELLS*)

1SDP-2 Native mass spectrometry of biomolecular complexes
Satoko Akashi (*Grad. Sch. Med. Life Science, Yokohama City Univ.*)

1SDP-3 (1Pos055) Biophysical analysis of alpha-synuclein oligomers by microchip electrophoresis
William E. Arter[1,2], Catherine K. Xu[1], Georg Krainer[1], Christopher M. Dobson[1], Tuomas P. J. Knowles[1,2] ([1]*Centre for Misfolding Disease, Department of Chemistry, University of Cambridge*, [2]*Cavendish Laboratory, Department of Physics, University of Cambridge*)

1SDP-4 (1Pos267) Visualization and quantification of biological samples by high-speed atomic force microscope
Hiroki Watanabe[1,2], Koichi Kato[1,2,3], Takayuki Uchihashi[1,4] ([1]*NINS, ExCELLS*, [2]*NINS, IMS*, [3]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [4]*Dept. Phys., Nagoya Univ.*)

1SDP-5 Dynamic structures and interactions of antibodies under physiologically relevant conditions
Saeko Yanaka[1,2,3], Rina Yogo[1,2,3], Hirokazu Yagi[3], Koichi Kato[1,2,3] ([1]*IMS., Natl. Inst. Nat. Sci.*, [2]*ExCELLS, Natl. Inst. Nat. Sci.*, [3]*Grad. Sch. Pharma. Sci., Nagoya City Univ.*)

1SDP-6 (1Pos058) 新規に開発した高濃度タンパク質のためのネガティブ染色電子顕微鏡法
(1Pos058) A newly developed negative stain EM method for protein complexes at high protein concentration
○今井 洋 [1], 加藤 貴之 [2], Christoph Gerle[3], 武藤 悦子 [4], 光岡 薫 [5], 栗栖 源嗣 [3], 難波 啓一 [2], 昆 隆英 [1]（[1]阪大・院理・生物科学, [2]阪大・生命機能, [3]阪大・蛋白研, [4]理研 CBS, [5]阪大・超高圧電顕センター）
Hiroshi Imai[1], Takayuki Kato[2], Gerle Christoph[3], Etsuko Muto[4], Kaoru Mitsuoka[5], Genji Kurisu[3], Keiichi Namba[2], Takahide Kon[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*IPR, Osaka Univ.*, [4]*CBS, RIKEN*, [5]*Res. Ctr. UVHEM, Osaka Univ.*)

1SDP-7 蛋白質相互作用の熱測定と創薬
Thermodynamics of Protein Interaction for Therapy and Diagnosis
長門石 曉 [1,2], ○津本 浩平 [1,2]（[1]東京大学医科学研究所, [2]東京大学大学院工学系研究科）
Satoru Nagatoishi[1,2], **Kohei Tsumoto**[1,2] ([1]*Inst Med Sci, Univ Tokyo*, [2]*Sch Eng, Univ Tokyo*)

おわりに
Closing Remarks

13:40〜16:20　E 会場（4F クリスタルルーム）／Room E (4F Crystal Room)
1SEP　共催：JST さきがけ「1 細胞解析」
さきがけ「1 細胞」は何をやっている？　1 細胞研究の醍醐味と技術革新
What is “Single-cell PRESTO” doing?

オーガナイザー：城口 克之（理化学研究所），鈴木 団（大阪大学）
Organizers: Katsuyuki Shiroguchi (RIKEN), Madoka Suzuki (Osaka University)

We, the researchers in the “Single-cell PRESTO” project, have heterogeneous research interests. In fact, our projects are diverse; imaging, (fluorescent) probes, gene expression, omics analysis, brain/neurons, membrane, sequencing and quantification of nucleic acid molecules, development, heat-sensing/manipulation, cell-measurement/manipulation/modeling, and glycans. However, we are gathered with a keyword “single-cell studies”. Then, what is it? In this symposium, selected members will guide you to the exciting “single-cell studies” by presenting the significance and advantages of their own single-cell studies, development of new techniques, and challenges for new research fields.

1SEP-1　Cellomics approach for high-throughput functional annotation of Caenorhabditis elegans neural network
Wataru Aoki[1,2], Yuji Yamauchi[1], Mitsuyoshi Ueda[1] ([1]*Graduate School of Agriculture, Kyoto University*, [2]*JST, PREST*)

1SEP-2　(1Pos196) Single-cell trajectory analysis of human iPS cell-derived neurons carrying a rare RELN deletion
Yuko Arioka[1,2,3], Emiko Shishido[1,4], Norio Ozaki[1] ([1]*Department of Psychiatry, Nagoya University Graduate School of Medicine*, [2]*Nagoya University Hospital*, [3]*Institute for Advanced Research, Nagoya University*, [4]*National Institute for Physiological Sciences*)

1SEP-3　Multiphoton imaging and photostimulation techniques by spatio-temporal control of excitation pulses
Keisuke Isobe, Katsumi Midorikawa (*RIKEN RAP*)

1SEP-4　Chemical probes for fluorescence imaging or ablation of lacZ-positive cells with single cell resolution
Mako Kamiya (*Grad. Sch. Med., Univ. Tokyo*)

1SEP-5　三次元バーテックスモデル：三次元多細胞動態の 1 細胞統合モデリング
3D vertex model: single cell-integrated modeling of multi-cellular dynamics in three-dimensions
○奥田 覚 [1,2]（[1]金沢大・ナノ研, [2]JST さきがけ）
Satoru Okuda[1,2] ([1]*Nano LSI, Kanazawa Univ*, [2]*JST PRESTO*)

1SEP-6　分子解像度での生命理解に向けて
Towards molecular-resolved biology
○谷口 雄一（理研・BDR）
Yuichi Taniguchi (*RIKEN BDR*)

1SEP-7　1 細胞操作のための光応答性細胞固定化剤の開発
Photo-responsive cell immobilization tools for single-cell manipulation
○山口 哲志 [1,2]（[1]東京大学先端科学技術研究センター, [2]JST さきがけ）
Satoshi Yamaguchi[1,2] ([1]*Research Center for Advanced Science and Technology (RCAST), The University of Tokyo*, [2]*PRESTO, JST*)

13:40〜16:20　F 会場（4F マーブルルーム）／Room F (4F Marble Room)

1SFP　高感度水素検出による生体内化学反応の制御を目指して
Toward the chemical reaction control in biological environment by high-sensitive hydrogen detection

オーガナイザー：田中 伊知朗（茨城大学），石北 央（東京大学）
Organizers: Ichiro Tanaka (Ibaraki University), Hisroshi Ishikita (The University of Tokyo)

Hydrogen atoms play a crucial role in biological systems, including enzymatic reactions, but the presence cannot be identified in the X-ray crystal structures. Next-generation neutron sources (e.g., J-PARC) enable us to carry out neutron diffraction analysis of proteins, which provides us a lot of useful information of hydrogen atoms. In this symposium, top researchers in the fields of structural analysis, theoretical analysis, and spectroscopic analysis of proteins and nano-devices will discuss and overview what can be elucidated by visualizing and characterizing hydrogen atoms.

はじめに
Opening Remarks

1SFP-1　**単結晶中性子回折計 iBIX の現状と将来展望**
Current status and future prospects of single-crystal neutron diffractometer iBIX at pulsed neutron source MLF, J-PARC
○日下 勝弘[1], 山田 太郎[1], 矢野 直峰[1], 細谷 孝明[1], 大原 高志[2], 田中 伊知朗[1]（[1]茨城大学，フロンティア応用原子科学研究センター，[2]日本原子力研究開発機構，J-PARC センター）
Kastuhiro Kusaka[1], Taro Yamada[1], Naomine Yano[1], Takaaki Hosoya[1], Takashi Ohhara[2], Ichiro Tanaka[1]
([1]*Frontier Researc Center for Applied Atomic Sciences, Ibaraki University*, [2]*Japan Atomic Energy Agency, J-PARC Center*)

1SFP-2　**中性子結晶構造解析で明らかになるセルラーゼの加水分解メカニズム**
Hydrolytic mechanisms of inverting cellulases clarified by neutron crystallography
○五十嵐 圭日子[1,2]（[1]東京大学, [2]VTT フィンランド技術研究センター）
Kiyohiko Igarashi[1,2] ([1]*University of Tokyo*, [2]*VTT Technical Research Centre of Finland*)

1SFP-3　**フェレドキシン依存性ビリン還元酵素の機能と基質のプロトン化状態**
Function of a ferredoxin-dependent bilin reductase and the protonation state of its substrate
○海野 昌喜[1,2]（[1]茨城大学大学院理工学研究科量子線科学専攻, [2]茨城大学フロンティア応用原子科学研究センター）
Masaki Unno[1,2] ([1]*Graduate School of Science & Engineering, Ibaraki University*, [2]*Frontier Research Center for Applied Atomic Sciences*)

1SFP-4　**(1Pos021) 創薬標的タンパク質の中性子結晶構造解析**
(1Pos021) Neutron crystallographic analysis of drug-target proteins
○横山 武司（富山大・薬）
Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

1SFP-5　Liquid properties and chemical reactions in 100 nm nanochannels
Kazuma Mawatari (*Univ. Tokyo*)

1SFP-6　**光合成水分解反応におけるプロトンおよび水分子の赤外分光検出**
Infrared detection of protons and water molecules in photosynthetic water oxidation
○野口 巧（名古屋大学大学院理学研究科物質理学専攻（物理系））
Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

おわりに
Closing Remarks

13:40〜16:20　G 会場（4F アイボリールーム）／Room G (4F Ivory Room)
1SGP　高次元データ駆動科学と計測インフォマティクスによる分子観察の新展開
New horizon in molecular observation through high-dimensional data-driven and measurement informatics approaches

オーガナイザー：木川 隆則（理化学研究所），松永 康佑（埼玉大学）
Organizers: Takanori Kigawa (RIKEN), Yasuhiro Matsunaga (Saitama University)

Recent intelligent measurement/analysis methods achieved by "measurement informatics" have improved resolutions and efficiencies of measurement technologies. On the other hand, we are at a loss for analyzing the vast amount of data generated through the development of those technologies. In order to promote the "high-dimensional data-driven science", which makes full use of statistics and computational technology to acquire scientific knowledge from the high-dimensional data, it is essential to establish logical data analysis and modeling methods. In this symposium, the recent advancements in statistical science, large-scale computing, and molecular measurement will be presented in order to understand the general concept of high-dimensional data-driven and measurement informatics approaches and discuss its application to life science.

1SGP-1　ポピュレーションアニーリングによるベイズ推定
Bayesian inference with population annealing
○福島 孝治（東京大学）
Koji Hukushima (*The University of Tokyo*)

1SGP-2　Objective and efficient procedure for inferring couplings in neuronal networks
Yu Terada[1,2], Tomoyuki Obuchi[1], Takuya Isomura[2], **Yoshiyuki Kabashima**[1] ([1]*Tokyo Tech.*, [2]*RIKEN CBS*)

1SGP-3　定量的安定同位体標識とテンソル分解による重複 NMR シグナルの分解法
Solving signal overlap in NMR spectra using quantitative isotope labeling and tensor decomposition
○葛西 卓磨 [1,2], 小野 峻佑 [2,3], 田中 利幸 [4], 池田 思朗 [5], 木川 隆則 [1,3]（[1]理研・生命機能, [2]JST・さきがけ, [3]東工大・情報理工, [4]京大院・情報, [5]統数研）
Takuma Kasai[1,2], Shunsuke Ono[2,3], Toshiyuki Tanaka[4], Shiro Ikeda[5], Takanori Kigawa[1,3] ([1]*RIKEN BDR*, [2]*PRESTO, JST*, [3]*Sch. Comput., Tokyo Inst. Tech.*, [4]*Grad. Sch. Inform., Kyoto Univ.*, [5]*Inst. Stat. Math.*)

1SGP-4　高速高精度一分子計測により明らかとなったキチン分解酵素の運動機構
Moving mechanism of chitin hydrolase was revealed by high precision and speed single molecule analysis
○中村 彰彦 [1,2], 岡崎 圭一 [1], 古田 忠臣 [3], 櫻井 実 [3], 飯野 亮太 [1,2]（[1]自然科学研究機構 分子科学研究所, [2]総合研究大学院大学, [3]東京工業大学）
Akihiko Nakamura[1,2], Kei-ichi Okazaki[1], Tadaomi Furuta[3], Minoru Sakurai[3], Ryota Iino[1,2] ([1]*Institute for Molecular Science*, [2]*SOKENDAI*, [3]*Tokyo Institute of Technology*)

1SGP-5　DNA curtains: high-throughput single molecule imaging for DNA transactions
Tsuyoshi Terakawa (*Grad. School of Sci., Kyoto Univ.*)

1SGP-6　2 次元／3 次元 AFM によるバイオ系試料の観察と高度な AFM データ解析の必要性
2D/3D-AFM imaging of biological systems and demands for advanced AFM data analysis
○福間 剛士（金沢大・ナノ生命研）
Takeshi Fukuma (*NanoLSI, Kanazawa Univ.*)

1SGP-7　細胞表面動態の高精度計測を目指した高速原子間力顕微鏡装置の開発
A High-speed atomic force microscope for detailed time series analysis of cell surface dynamics
○清水 将裕[1,2], 岡本 千優[3], 今井 大達[1], 渡辺 信嗣[4], 安藤 敏夫[2,4], 古寺 哲幸[2,4]（[1]金沢大・新学術創成, [2]JST・CREST, [3]金沢大・数物, [4]金沢大・WPI-NanoLSI）
Masahiro Shimizu[1,2], Chihiro Okamoto[3], Hirotatsu Imai[1], Shinji Watanabe[4], Toshio Ando[2,4], Noriyuki Kodera[2,4] ([1]*InFiniti, Kanazawa Univ.*, [2]*CREST, JST*, [3]*Dept. Sch. Math. & Phys., Kanazawa Univ.*, [4]*WPI-NanoLSI, Kanazawa Univ.*)

1SGP-8　高速原子間力顕微鏡 1 分子計測と分子シミュレーションのデータ同化による生体分子 4 次元構造解析
Four-dimensional biomolecular structure analysis with data assimilation of HS-AFM single molecule measurement and molecular simulation
○渕上 壮太郎[1,2], 新稲 亮[1], 高田 彰二[1,2]（[1]京大院・理, [2]JST・CREST）
Sotaro Fuchigami[1,2], Toru Niina[1], Shoji Takada[1,2] ([1]*Grad. Sch. of Science, Kyoto Univ.*, [2]*CREST, JST*)

13:40〜16:20　H 会場（4F アンバールーム）／Room H (4F Amber Room)
1SHP　GPCR による多様な情報伝達機構を解き明かす構造―機能相関研究の新展開
Frontier of structure-function studies to unveil diverse GPCR signaling

オーガナイザー：片山 耕大（名古屋工業大学），寿野 良二（関西医科大学）
Organizers: Kota Katayama (Nagoya Institute of Technology), Ryoji Suno (Kansai Medical University)

GPCR signaling utilizes an allosteric coupling between the extracellular facing ligand-binding pocket and the cytoplasmic domain of the receptor selectively interacting with signal transducer. This allosteric effect enables one site of the receptor to regulate the function of another spatially distinct region. Therefore, it is important to understand the molecular mechanisms behind ligand-induced changes in receptor conformation and specific transducer-recognition for the development of GPCR-based drugs. This symposium is dedicated to discuss the latest trends on the structure-function studies to explore the molecular basis of GPCR signal transduction.

1SHP-1　Holistic Phenotyping of GPCR Signaling System by a Versatile Single-Platform Assay
Ikuo Masuho (*The Scripps Research Institute FL, Department of Neuroscience*)

1SHP-2　GPCR の 1 分子拡散動態から複数の薬効を読み解く
Estimation of multiple drug effects on GPCR based on the single-molecule diffusion dynamics
○柳川 正隆（理研・佐甲細胞情報研究室）
Masataka Yanagawa (*Cellular Informatics Lab., Riken*)

1SHP-3　Structure and conformational transitions of a neurotensin receptor 1 Gi1 protein complex
Hideaki Kato[1,3], Yan Zhang[2,3], Hongli Hu[3], Carl-Mikael Suomivuori[3], Francois Marie Ngako Kadji[4], Junken Aoki[4], Kaavya Krishna Kumar[3], Rasmus Fonseca[3], Daniel Hilger[3], Weijiao Huang[3], Naomi Latorraca[3], Asuka Inoue[4], Ron Dror[3], Brian Kobilka[3], Georgios Skiniotis[3] ([1]*The Univ. of Tokyo*, [2]*Zhejiang Univ.*, [3]*Stanford Univ.*, [4]*Tohoku Univ.*)

1SHP-4 NMR 法を用いた動的構造にもとづく GPCR のシグナル伝達機構の解明
Dynamics of G protein-coupled receptor related to various signaling revealed by NMR
○幸福 裕[1], 白石 勇太郎[1], 夏目 芽衣[1], 奥出 順也[1], 今井 駿輔[1], 前田 正洋[2], 辻下 英樹[2], 倉永 健史[1], 井上 将行[1], 中田 國夫[3], 水越 利巳[3], 上田 卓見[1], 岩井 秀夫[4], 嶋田 一夫[1]（[1]東大・院薬系, [2]塩野義製薬（株）, [3]味の素（株）, [4]ヘルシンキ大）
Yutaka Kofuku[1], Yutaro Shiraishi[1], Mei Natsume[1], Junya Okude[1], Shunsuke Imai[1], Masahiro Maeda[2], Hideki Tsujishita[2], Takefumi Kuranaga[1], Masayuki Inoue[1], Kunio Nakata[3], Toshimi Mizukoshi[3], Takumi Ueda[1], Hideo Iwai[4], Ichiro Shimada[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*Shionogi Co., Ltd.*, [3]*Ajinomoto Co., Inc.*, [4]*Univ. Helsinki*)

1SHP-5 Theoretical Prediction of Thermostabilizing Mutations for GPCR: Identification of Hot-Spot Residues to be Mutated Common in Class A GPCRs
Satoshi Yasuda[1,2,3], Yuta Kajiwara[4], Yuki Takamuku[1], Nanao Suzuki[1], Yosuke Toyoda[5], Kazushi Morimoto[5], Ryoji Suno[5], So Iwata[5], Takuya Kobayashi[5], Takeshi Murata[1,2], Masahiro Kinoshita[3] ([1]*Grad. Sch. Sci., Chiba Univ.*, [2]*MCRC*, [3]*IAE*, [4]*Grad. Sch. Ener. Sci., Kyoto Univ.*, [5]*Grad. Sch. Med., Kyoto Univ.*)

1SHP-6 (1Pos077) 理論計算による熱安定化ムスカリン M2 受容体の選択的アンタゴニスト AF-DX 384 結合型構造
(1Pos077) Structural insights into the subtype-selective antagonist binding to the M2 muscarinic receptor
○寿野 良二[1], Lee Sangbae[2], 前田 将司[3], 安田 賢司[4], 山下 恵太郎[9], 平田 邦生[5,6], 村田 武士[7], 木下 正弘[8], 山本 雅貴[5], Kobilka Brian[3], Vaidehi Nagarajan[2], 岩田 想[8], 小林 拓也[1]（[1]関西医大・医, [2]ホープ市医学センター, [3]スタンフォード大・医, [4]千葉大・理, [5]理研・SPring-8, [6]JST・さきがけ, [7]京大・エネ研, [8]京大・医, [9]東大・理）
Ryoji Suno[1], Sangbae Lee[2], Shoji Maeda[3], Satoshi Yasuda[4], Keitaro Yamashita[9], Kunio Hirata[5,6], Takeshi Murata[7], Masahiro Kinoshita[8], Masaki Yamamoto[5], Brian Kobilka[3], Nagarajan Vaidehi[2], So Iwata[8], Takuya Kobayashi[1] ([1]*Kansai Medi. Univ.*, [2]*City Hope Med. Ctr.*, [3]*Stanford Univ.*, [4]*Chiba Univ.*, [5]*RIKEN,SPring-8*, [6]*JST,PRESTO*, [7]*IAE, Kyoto Univ.*, [8]*Med,Kyoto Univ.*, [9]*Univ.Tokyo,Sci*)

2 日目（9 月 25 日（水））／Day 2（Sep. 25 Wed.）

8:30〜11:10　B 会場（4F 天玉）／Room B (4F Tengyoku)
2SBA　共催：新学術領域研究「分子夾雑の生命化学」
分子夾雑のススメ
Invitation to multimolecular crowding

オーガナイザー：田端 和仁（東京大学），三好 大輔（甲南大学）
Organizers: Kazuhito Tabata (The University of Tokyo), Daisuke Miyoshi (Konan University)

The inside of the cell is occupied by a wide variety of different biomolecules, and the localization and concentration of the biomolecules change dynamically depending on the cell cycle and state. Since the begining of life, biomolecules have evolved to express functions in multi-molecular crowding environments. However, attempts to elucidate the nature of multi-molecular crowding environment have just begun. In this symposium, we focus on liquid-liquid phase separation phenomena that are related to multi-molecular crowding, and discuss the current topics and prospects of multi-molecular crowding research.

2SBA-1 Chemical control of protein localization in the multimolecular cellular space
Shinya Tsukiji (*Grad. Sch. Eng., Nagoya Inst. Tech.*)

2SBA-2 Secondary structure of DNA for liquid-liquid phase separation
Masahiro Mimura[1,2], Shunsuke Tomita[2], Ryoji Kurita[1,2], Kentaro Shiraki[1] ([1]*Pure and Appl. Sci., Univ. Tsukuba*, [2]*Biomed. Res. Inst., AIST.*)

2SBA-3 **相分離生物学：相分離する LC ドメイン**
Phasing Biology: Low-Complexity Domains Phase-Separate through Cross-β Interaction
○森 英一朗（奈良県立医科大学 医学部 未来基礎医学）
Eiichiro Mori (*Dept. Future Basic Med., Sch. Med., Nara Med. Univ.*)

2SBA-4 **分子夾雑系における光センサー蛋白質の動的挙動―揺らぎと反応ダイナミクス―**
Fluctuation and reaction dynamics of a light sensor protein in crowding environment
○中曽根 祐介, 村上 大斗, 寺嶋 正秀（京大・院理）
Yusuke Nakasone, Hiroto Murakami, Masahide Terazima (*Grad. Sch. Sci., Kyoto Univ.*)

2SBA-5 **細胞様構造や細胞組織体の自己創生：分子夾雑系の活用**
Self-emergence of primitive cell and cellular mini-organoids under crowding environment
○吉川 研一（同志社大学 生命医科学部）
Kenichi Yoshikawa (*Facul. Life Med. Sci., Doshisha Univ.*)

8:30～11:10　C 会場（4F 天樹）／Room C (4F Tenjyu)
2SCA　分子構造ビッグデータの時代のバイオインフォマティクスの挑戦
Challenges of bioinformatics for the era of molecular structure big-data

オーガナイザー：白井 剛（長浜バイオ大学），寺田 透（東京大学）
Organizers: Tsuyoshi Shirai (Nagahama Institute of Bioscience and Technology), Tohru Terada (The University of Tokyo)

The recent rapid developments in the techniques of supramolecular structure and high-throughput omics analyses enhanced the importance of the bioinformatics/data science to analyze and mining knowledge from “molecular structure big-data”. This symposium will be focused on reviewing and discussing the recent researches in this field produced from AMED-BINDS activities.

2SCA-1 AMED-BINDS 事業におけるビッグデータ科学
Big Data Science at AMED-BINDS
○中村 春木（大阪大学蛋白質研究所）
Haruki Nakamura (*Institute for Protein Research*)

2SCA-2 **多層ニューラルネットワークを用いたタンパク質残基間コンタクトおよびタンパク質 - 基質相互作用の予測**
Prediction of protein residue contacts and protein-ligand interactions with deep neural networks
○富井 健太郎（産業技術総合研究所）
Kentaro Tomii (*National Institute of Advanced Industrial Science and Technology (AIST)*)

2SCA-3 Integrated approach of experimental data and computer modeling and simulation for understanding chromatin structure and dynamics
Hidetoshi Kono, Atsushi Matsumoto, Shun Sakuraba, Hisashi Ishida (*QST, Institute for Quantum Life Science (iQLS), MMS*)

2SCA-4 電顕のインフォマティクス：2D 生画像データの収集と原子モデルのフィッティング
EM informatics: archiving raw 2D images and fitting atomic models into a map
○川端 猛, 栗栖 源嗣（大阪大学　蛋白質研究所）
Takeshi Kawabata, Genji Kurisu (*Inst.Prot.Res., Osaka Univ.*)

2SCA-5 Development of a deep-learning-based method to identify "good" regions of a cryo-EM grid
Tohru Terada[1,3], Yuichi Yokoyama[2], Kentaro Shimizu[1,3], Kouki Nishikawa[4], Yoshinori Fujiyoshi[4], Kazutoshi Tani[5] ([1]*III, Univ. Tokyo*, [2]*GSII, Univ. Tokyo*, [3]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [4]*Adv. Res. Inst., Tokyo Med. Dent. Univ.*, [5]*Grad. Sch. Med., Mie Univ.*)

2SCA-6 クライオ電顕データ収集の効率化に資する凍結グリッド作成法やソフトウェア
Improvements in grid preparation method and software for facilitating cryoEM data collection
○難波 啓一 [1,2,3]（[1]大阪大学大学院生命機能研究科, [2]理研放射光科学研究センター, [3]理研生命機能科学研究センター）
Keiichi Namba[1,2,3] ([1]*Graduate School of Frontier Biosciences, Osaka University*, [2]*RIKEN SPring-8 Center*, [3]*RIKEN Center for Biosystems Dynamics Research*)

8:30〜11:10　D 会場（4F 天葉）／Room D (4F Tenyo)

2SDA　共催：新学術領域研究「発動分子科学：エネルギー変換が拓く自律的機能の設計」 生体分子機械の非平衡エナジェティクス
Nonequilibrium Energetics of Biological Molecular Machines

オーガナイザー：Chun-Biu Li（Stockholm University），鳥谷部 祥一（東北大学）
Organizers: Chun-Biu Li (Stockholm University), Shoichi Toyabe (Tohoku University)

Many bio-molecular motors can operate robustly and energetic efficiently in the highly fluctuating nano-scale. How these molecules achieve such remarkable functions is an intriguing question that requires the understanding of the general principles of structure and design, enzymatic kinetics and nonequilibrium physics of biological machineries. By bringing together both experimental and theoretical experts from interdisciplinary fields, this symposium aims to explore A) Novel experimental techniques to probe the energetic efficiency in the single molecule level; B) New theoretical methods to explain the biophysical principles of molecular energetics; C) Common strategies shared among different bio-molecular machines in achieving energetic efficiency.

2SDA-1 生物系のエナジェティクス
Energetics of biological system
○鳥谷部 祥一（東北大・院工）
Shoichi Toyabe (*Grad. Sch. Eng., Tohoku Univ.*)

2SDA-2 歩行型モーター・キネシン 1 の非平衡エネルギー論
Nonequilibrium Energetics of a walking motor kinesin-1
○有賀 隆行（山口大学医学系研究科）
Takayuki Ariga (*Graduate school of medicine, Yamaguchi University*)

2SDA-3 FoF1-ATP 合成酵素の回転力はどのようにして発生しているのか? 構造生物・生物物理学的手法による解析
Structural and biophysical analyses of torque generation mechanism of F_oF_1-ATP synthase
○鈴木 俊治（東京工業大学 科学技術創成研究院 化学生命科学研究所）
Toshiharu Suzuki (*Lab for Chem and Life Sci, Inst of Innov Res, Tokyo Inst of Tech*)

2SDA-4 Modeling of myosin V motor dynamics to understand high-speed AFM observations
Holger Flechsig (*WPI Nano Life Science Institute, Kanazawa University*)

2SDA-5 Energetics and structural dynamics of a viral RNA polymerase ratcheting along DNA with fidelity control
Jin Yu (*Beijing Computational Science Research Center*)

2SDA-6 Error-speed correlations in biopolymer synthesis
Simone Pigolotti (*Okinawa Institute of Science and Technology Graduate University*)

2SDA-7 (2Pos121) Dynamic energy landscape of a linear motor chitinase from single-particle tracking trajectories
Kei-ichi Okazaki, Akihiko Nakamura, Ryota Iino (*Institute for Molecular Science*)

8:30〜11:10　E 会場（4F クリスタルルーム）／Room E (4F Crystal Room)
2SEA　放射光利用生物物理研究の最前線
Frontiers of Synchrotron Radiation Biophysics

オーガナイザー：岩本 裕之（高輝度光科学研究センター），関口 博史（高輝度光科学研究センター）
Organizers: Hiroyuki Iwamoto (JASRI), Hiroshi Sekiguchi (JASRI)

Synchrotron radiation X-rays has a wide range of applications for life sciences, including fiber diffraction and protein solution scattering, besides the most popular protein crystallography. This symposium sheds lights on synchrotron radiation techniques other than protein crystallography, and introduce the recent progress of these techniques and the results of the latest research.

はじめに
Opening Remarks
岩本 裕之（高輝度光科学研究センター）
Hiroyuki Iwamoto (*JASRI*)

2SEA-1 非結晶生体試料のシンクロトロン放射光 X 線回折実験に関する最近の進歩
Recent progress in synchrotron radiation X-ray diffraction studies for non-crystalline biological specimens
○岩本 裕之（SPring-8・JASRI）
Hiroyuki Iwamoto (*SPring-8, JASRI*)

2SEA-2 X 線繊維回折法によって明らかとなった秒単位の微小管構造変化
Dynamic changes of tubulin dimer configurations on a scale of sub-second revealed by high flux X-ray fiber diffraction
○上村 慎治 [1], 今井 洋 [2], 八木 俊樹 [3], 岩本 裕之 [4]（[1]中大・理工・生命, [2]阪大・院理・生物科学, [3]県立広島大・生命環境, [4]SPring-8, JASRI）
Shinji Kamimura[1], Hiroshi Imai[2], Toshiki Yagi[3], Hiroyuki Iwamoto[4] ([1]*Dept. Biol. Sci., Chuo Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Dept. Life Sci., Prefect. Univ. Hiroshima*, [4]*SPring-8, JASRI*)

2SEA-3 Diffracted X-ray Tracking for protein dynamics
Hiroshi Sekiguchi (*Cent. Synchrotron Rad. Res., JASRI/SPring-8*)

2SEA-4 Dynamic changes in cardiac myosin head regulation during hyperglycemic events in insulin resistant rats
James T. Pearson[1,2], Naoto Yagi[3], Mikiyasu Shirai[1], Mark Waddingham[1], Hirotsugu Tsuchimochi[1], Takashi Sonobe[1], Vijayakumar Sukumaran[1] ([1]*National Cerebral and Cardiovascular Center*, [2]*Monash University, Department of Physiology*, [3]*JASRI*)

2SEA-5 (2Pos269) G1 期酵母細胞核内における核酸分布の XFELX 線回折イメージング
(2Pos269) Distribution of nucleic acids in yeast nucleus of G1 phase visualized by X-ray diffraction imaging using X-ray free electron laser
○中迫 雅由 [1,2], 山本 隆寛 [1,2], 小林 周 [1,2], 大出 真央 [1,2], 岡島 公司 [1,2], 高山 裕貴 [1,2,3], 苣口 友隆 [1,2], 山本 雅貴 [2]（[1]慶應義塾大学, [2]理化学研究所, [3]兵庫県立大）
Masayoshi Nakasako[1,2], Takahiro Yamamoto[1,2], Amane Kobayashi[1,2], Mao Oide[1,2], Koji Okajima[1,2], Yuki Takayama[1,2,3], Tomotaka Oroguchi[1,2], Masaki Yamamoto[2] ([1]*Keio University*, [2]*RIKEN*, [3]*University of Hyogo Prefecture*)

おわりに
Closing Remarks
岩本 裕之（高輝度光科学研究センター）
Hiroyuki Iwamoto (*JASRI*)

8:30〜11:10　F 会場（4F マーブルルーム）／Room F (4F Marble Room)
2SFA　共催：JST さきがけ「生命機能メカニズム解明のための光操作技術」
光操作による生命機能解析
Elucidation of biological functions by optical control

オーガナイザー：七田 芳則（立命館大学），塚本 寿夫（分子科学研究所）
Organizers: Yoshinori Shichida (Ritsumeikan University), Hisao Tsukamoto (Institute for Molecular Science)

Life science has dramatically progressed through the development of powerful biophysical techniques controlling cellular functions by light. For example, Optogenetics utilizing photoreceptive proteins as an optical controlling tool has been established to analyze and manipulate various biological functions. In this symposium, active young researchers in biology and engineering will present how to elucidate biological functions by these techniques, and we and participants will discuss direction of the research in this field including necessary biophysical tools to be developed.

はじめに
Opening Remarks
七田 芳則（立命大）
Yoshinori Shichida (*Ritsumeikan Univ.*)

2SFA-1 記憶痕跡サブ・アンサンブルの協奏的活動によるエピソード記憶の脳内表現
Orchestrated ensemble activities constitute a hippocampal memory engram
○大川 宜昭 [1,2]（[1]富山大学・院医薬, [2]JST・さきがけ）
Noriaki Ohkawa[1,2] ([1]*Univ of Toyama Grad Sch of Med and Pharm Sci*, [2]*PRESTO, JST*)

2SFA-2 X 線を用いた神経機能の遠隔無線操作
Remote and wireless control of neuronal function using X-ray
○山下 貴之 [1]（[1]名古屋大学 環境医学研究所 神経系分野 2, [2]科学技術振興機構 さきがけ）
Takayuki Yamashita[1] ([1]*Dept. Neurosci. II, RIEM, Nagoya Univ.*, [2]*PRESTO, JST*)

2SFA-3 バッテリレス超小型光刺激デバイス
Batteryless ultra-small implantable optical stimulator
○德田 崇 [1], Pakpuwadon Thanet[2], Wuthibenjaphonchai Nattakarn[2], 春田 牧人 [2], 笹川 清隆 [2], 太田 淳 [2]（[1]東工大 未来研, [2]奈良先端大 物質創成）
Takashi Tokuda[1], Thanet Pakpuwadon[2], Nattakarn Wuthibenjaphonchai[2], Makito Haruta[2], Kiyotaka Sasagawa[2], Jun Ohta[2] ([1]*FIRST, Tokyo Tech*, [2]*Mater. Sci., NAIST*)

2SFA-4　長波長レーザーによる超深部イメージングシステムの開発
Development of deep-tissue imaging system based on a long-wavelength laser
○野村 雄高（分子科学研究所）
Yutaka Nomura (*Institute for Molecular Science*)

2SFA-5　光操作型アデノウイルスベクターの開発と応用
Generation of photoactivatable adenovirus vector for spatiotemporally controllable gene therapy
○高山 和雄 [1,2,3]（[1]阪大院薬, [2]医薬基盤健栄研, [3]さきがけ）
Kazuo Takayama[1,2,3] ([1]*Osaka University*, [2]*NIBIOHN*, [3]*PRESTO*)

2SFA-6　(2Pos198) 微生物型ロドプシンに基づく光遺伝学ツールの探索と開発
(2Pos198) Exploration and development of microbial rhodopsin-based optogenetic tools
○小島 慧一, 須藤 雄気（岡山大・院・医歯薬(薬)）
Keiichi Kojima, Yuki Sudo (*Grad. Sch. of Med. Dent. Pharm. Sci., Okayama Univ.*)

2SFA-7　「総力戦」としての光操作技術
Optical control as a fusion of neuroscience, medicine, engineering, and biophysics
○塚本 寿夫 [1,2]（[1]分子科学研究所, [2]JST さきがけ）
Hisao Tsukamoto[1,2] ([1]*Institute for Molecular Science*, [2]*PRESTO, JST*)

8:30～11:10　G 会場（4F アイボリールーム）／Room G (4F Ivory Room)
2SGA　電子・熱・化学エネルギーの生体内伝達と地域社会実装に向けた基盤研究
How is 'ENERGY' generated/transferred across the cellular systems?

オーガナイザー：和田 啓（宮崎大学），榊原 陽一（宮崎大学）
Organizers: Kei Wada (University of Miyazaki), Yoichi Sakakibara (University of Miyazaki)

The cellular systems involved in the generation/transfer of ENERGY are 'awesome'. For instance, ATP is known in biochemistry as the "molecular currency" of intracellular energy transfer; that is, ATP is able to store and transport chemical energy within cells. The cells also possess the sophisticated systems including the utilizing the reactive sulfur species and the transfer mechanism of the electrons and the thermal energy. This symposium covers a variety of topics regarding the recent findings and advances in the biochemistry/chemistry of the energy related compounds.

2SGA-1　生体内の電子伝達金属補因子「鉄硫黄クラスター」の生合成機構
Molecular mechanism of the biosynthesis of the iron-sulfur clusters involved in the electron transport in vivo
○和田 啓（宮崎大 医）
Kei Wada (*Dept. of Medical Sciences, Univ. of Miyazaki*)

2SGA-2　高エネルギー硫酸ヌクレオチド分子を利用した生体内代謝：硫酸転移酵素の多様な生理機能の解明
Metabolism of key endogenous molecules mediated by sulfotransferases with a hi-energy sulfonucleotide, PAPS
○黒木 勝久 [1], 寺本 岳大 [2], 角田 佳充 [2], Liu Ming-Cheh[3], 水光 正仁 [1], 榊原 陽一 [1]（[1]宮崎大・農・応生科, [2]九大院・農・生命機能, [3]トレド大・薬）
Katsuhisa Kurogi[1], Takamasa Teramoto[2], Yoshimitsu Kakuta[2], Ming-Cheh Liu[3], Masahito Suiko[1], Yoichi Sakakibara[1] ([1]*Dept. Biochem. Appl. Biosci., Fac. Agric., Univ. Miyazaki*, [2]*Dept. Biosci. Biotechnol., Grad. Sch. Agric., Kyushu Univ.*, [3]*Dept. Pharmacol., Coll. Pharm., Univ. Toledo*)

2SGA-3 **発熱植物の生体エネルギー論：シアン耐性呼吸からミトコンドリアの構造まで**
Towards understanding the roles of mitochondrial energy bypasses in heat-producing plants
○稲葉 靖子（宮崎大・農）
Yasuko Ito-Inaba (*Agric, Univ. Miyazaki*)

2SGA-4 The role of Fhod family formin proteins in mouse heart
Fumiyuki Sanematsu[1], Hideki Sumimoto[2], Ryu Takeya[1] ([1]*Dept. of Pharmacol., Fac. of Med., Univ. of Miyazaki*, [2]*Dept. of Biochem., Kyushu Univ. Grad. Sch. of Med. Sci.*)

2SGA-5 In-cell ^{19}F NMR: Chemical method for investigating nucleic acid structure in living cells
Takumi Ishizuka, Yan Xu (*Fac. Med., Univ. of Miyazaki*)

2SGA-6 **微生物電池における c 型シトクロムを介した細胞外電子伝達**
Extracellular electron transfer via c-type cytochromes in microbial fuel cells
○井上 謙吾（宮崎大・農）
Kengo Inoue (*Department of Biochemistry and Applied Biosciences, Faculty of Agriculture*)

8:30〜11:10　H 会場（4F アンバールーム）／Room H (4F Amber Room)
2SHA　**タンパク質のダイナミックレスポンスに関わる未解決問題への挑戦**
Challenges to get insight into unsolved problems of dynamic response in proteins

オーガナイザー：鷹野 優（広島市立大学），米澤 康滋（近畿大学）
Organizers: Yu Takano (Hiroshima City University), Yasushige Yonezawa (Kindai University)

Recent studies have shown that Proteins are not static molecular machine, but play their biological role by dynamical response according to external perturbations. At present, it is believed the mechanical origin of the dynamical response involves in structure of proteins, dealing with varieties of biological systems. However, the mechanisms of the dynamic response is still debates. In this symposium, we will overlook the issue by presentations of researchers who have tackled to the problems using advanced technique or novel ideas, and discuss the future progress of this exciting scientific area.

はじめに
Opening Remarks
米澤 康滋（近畿大）
Yasushige Yonezawa (*Kindai Univ.*)

2SHA-1 **生物学的ネットワークの確率論的変動**
Stochastic usage of biological network
○白木 琢磨（近畿大学・生物理工学部）
Takuma Shiraki (*Kindai Univ. BOST*)

2SHA-2 Autoencoder-based analyses of dynamic allostery on proteins by regulator binding
Yuko Tsuchiya[1], Kei Taneishi[2], Yasushige Yonezawa[3] ([1]*AIRC, AIST*, [2]*RIKEN*, [3]*KINDAI University*)

2SHA-3 **大規模量子分子動力学シミュレーションを用いた光受容タンパク質におけるプロトン移動反応の解明**
Clarification of proton transfer reactions in photoreceptive proteins using large-scale quantum molecular dynamics simulations
○小野 純一[1], 岡田 千果[2], 西村 好史[1], 中井 浩巳[1,2,3]（[1]早大理工総研, [2]早大先進理工, [3]京大 ESICB）
Junichi Ono[1], Chika Okada[2], Yoshifumi Nishimura[1], Hiromi Nakai[1,2,3] ([1]*WISE, Waseda Univ.*, [2]*Grad. Sch. of Adv. Sci. & Eng., Waseda Univ.*, [3]*ESICB, Kyoto Univ.*)

2SHA-4 カルシウムシグナル伝達蛋白質 Calmodulin と結合ドメインの構造変化と相互作用
Conformational changes and interactions of calcium ion signal transfer protein Calmodulin and Calmodulin-binding domain
○下山 紘充（北里大学薬学部 生物分子設計学教室）
Hiromitsu Shimoyama (*Kitasato-Univ.*)

2SHA-5 タンパク質中の不均一なエネルギー流と機能に関する理論的研究
Theoretical study on non-uniform energy flow and protein function
○窪田 源己, Laprevote Olivier, 倭 剛久（名古屋大学）
Genki Kubota, Olivier Laprevote, Takahisa Yamato (*Nagoya University*)

2SHA-6 (2Pos057) ダイナミン GTP アーゼはアクチン線維の束化と分散を機械的に制御する
(2Pos057) Dynamin GTPase mechanically regulates bundling and unbundling of actin filaments
○竹居 孝二[1], テ モン ラ[1], 阿部 匡[1], 竹田 哲也[1], 藤原 郁子[2], 成田 哲博[3]（[1]岡山大 院医歯薬, [2]大阪市大 院理 細胞機能, [3]名大 院理 構造生物学研究センター）
Kohji Takei[1], La The Mon[1], Tadashi Abe[1], Tetsuya Takeda[1], Ikuko Fujiwara[2], Akihiro Narita[3] ([1]*Grad.Sch. Med.Dent. Pharm.Sci., Okayama Univ.*, [2]*Dept. Biol.Facul.Sci., Osaka City Univ.*, [3]*Struct. Biol. Res. Ctr and Divi. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

2SHA-7 Analysis of Effect of Mutation on the Response for Membrane Depolarization in the Voltage-Gated Potassium Channel Kv1.2
Hiroko X. Kondo[1], Norio Yoshida[2], Gen Masumoto[3], Matsuyuki Shirota[4,5,6], Yu Takano[7], Kengo Kinoshita[5,6] ([1]*Fac. Eng., Kitami Inst. Tech.*, [2]*Grad. Sch. Sci., Kyushu Univ*, [3]*RIKEN ISC*, [4]*Grad. Sch. Med., Tohoku Univ.*, [5]*GSIS, Tohoku Univ.*, [6]*ToMMo, Tohoku Univ.*, [7]*Grad. Sch. Info. Sci., Hiroshima City Univ.*)

おわりに
Closing Remarks
鷹野 優（広島市大）
Yu Takano (*Hiroshima City Univ.*)

14:10〜16:50　B 会場（4F 天玉）／Room B (4F Tengyoku)
2SBP　共催：新学術領域研究「代謝統合オミクス」
読む×解く、代謝のアダプテーション
Measure x Analyze Metabolic Adaptation of Biological Systems

オーガナイザー：岡田 眞里子（大阪大学），馬場 健史（九州大学）
Organizers: Mariko Okada (Osaka University), Takeshi Bamba (Kyushu University)

The life system is maintained by dynamic tuning of metabolisms. Rewiring of the metabolic networks in bacteria, plants or human diseases is considered to be the results of the adaptation of their whole-body metabolisms to environment. The molecular mechanism underlying the metabolic adaptation can be only understood through measuring and analyzing "trans-omic" network, consisting of interactions among molecules across multi-omic layers, such as genome, transcriptome, proteome, and metabolome. Here we hold this symposium to shed light on strategies and obstacles in integrating multiple omic layers to establish trans-omic approaches, and to have discussions with cutting edge researchers in omics research fields.

2SBP-1 トランスオミクスに資する次世代メタボローム分析技術の開発
Development of next generation metabolome analytical technologies for trans-omics
○馬場 健史（九大・生医研）
Takeshi Bamba (*Med. Inst. Bioreg., Kyushu Univ.*)

2SBP-2 Novel sequencing methods to read the epigenome
Takashi Ito (*Kyushu Univ. Grad. Sch. Med. Sci.*)

2SBP-3 Sulfur Metabolism Rewiring in NRF2-Addicted Cancer Cells
Hozumi Motohashi (*Institute of Development, Aging and Cancer, Tohoku University*)

2SBP-4 条件付き独立性を用いたネットワーク構造推定
Network structure inference by conditional independence
○宇田 新介（九州大学）
Shinsuke Uda (*Kyushu university*)

2SBP-5 一細胞遺伝子発現プロファイルからの細胞集団および細胞間コミュニケーションの動態モデリング
Modeling dynamics of cell population and cell-to-cell communication from single-cell gene expression profiles
○島村 徹平（名古屋大学）
Teppei Shimamura (*Nagoya University*)

2SBP-6 オミクスを数理モデルでつなぐ
Analyze Omics data using kinetic model
○岡田 眞里子 [1]（[1]大阪大学蛋白質研究所細胞システム研究室, [2]理化学研究所 IMS）
Mariko Okada[1] ([1]*Institute for Protein Research, Osaka University*, [2]*RIKEN IMS*)

14:10〜16:50　C 会場（4F 天樹）／Room C (4F Tenjyu)
2SCP　共催：新学術領域研究「シンギュラリティ生物学」
シンギュラリティ生物学：少数の要素が全体の機能を変革する
Singularity Biology: Small elements change the function of the whole systems

オーガナイザー：小松崎 民樹（北海道大学），堀川 一樹（徳島大学）
Organizers: Tamiki Komatsuzaki (Hokkaido University), Kazuki Horikawa (Tokushima University)

In the field of biological science, discontinuous critical phenomena (singularities) are broadly seen, for example, the emergence of life from the primordial soup, or the evolution and outbreak of diseases. It has been indicated that only a small number of core elements are required to bring about discontinuous changes to an entire multi-component system. However, the mechanism-of-action that generates such singularity phenomena is not yet certain. For this aim, to develop an imaging platform that will achieve both wide field-of-view high-resolution imaging and high-speed long-term imaging and information analysis methods are highly desired. In this symposium, we are aimed at exploring possible biological subjects and the associated technological developments toward uncovering the underlying mechanisms for the generation of singularity cells as well as their biological functions.

はじめに
Opening Remarks
永井 健治（大阪大）
Takeharu Nagai (*Osaka Univ.*)

2SCP-1 アルツハイマー病の解明のためにシンギュラリティ生物学ができること
What singularity biology can do for understanding Alzheimer's disease
○坂内 博子 [1,2], 廣島 通夫 [3], 添田 義行 [4], 高島 明彦 [4]（[1]慶應大・医, [2]JST ERATO, [3]理研・BDR, [4]学習院大・理）
Hiroko Bannai[1,2], Michio Hiroshima[3], Yoshiyuki Soeda[4], Akihiko Takashima[4] ([1]*Keio Univ. Sch. Med.*, [2]*JST ERATO*, [3]*RIKEN BDR*, [4]*Gakushuin Univ, Faculty. Sci.*)

2SCP-2 Delineation of the activation trajectory of autoreactive T cells
Taku Okazaki, Hikari Okamura, Il-mi Okazaki, Kenji Shimizu, Takumi Maruhashi, Daisuke Sugiura (*Div Imm Reg, Inst Adv Med Sci, Tokushima Univ*)

2SCP-3 全脳イメージングシステム FAST を用いたアンバイアスで仮説に依らない脳内シンギュラリティの検出
Unbiased and hypothesis-free approach to detect singularity in the brain using whole-brain imaging system FAST
○橋本 均[1,2,3,4], 中澤 敬信[1,5], 勢力 薫[1,6], 笠井 淳司[1]（[1]大阪大・薬・神経薬理, [2]大阪大・連合小児発達・子どものこころの発達研究センター, [3]大阪大・データビリティフロンティア機構・バイオサイエンス部門, [4]大阪大・先導的学際研究機構・超次元ライフイメージング研究部門, [5]大阪大・歯・薬理, [6]大阪大・国際共創大学院学位プログラム推進機構）
Hitoshi Hashimoto[1,2,3,4], Takanobu Nakazawa[1,5], Kaoru Seiriki[1,6], Atsushi Kasai[1] ([1]*Lab. of Mol. Neuropharmacol., Grad. Sch. of Pharmaceut. Sci., Osaka Univ.*, [2]*Center for Child Mental Dev, United Grad. Sch. of Child Dev., Osaka Univ.*, [3]*Div. of Biosci., Inst. for Datability Sci., Osaka Univ.*, [4]*Transdimensional Life Imaging Div., Inst. for Open and Transdisciplinary Res. Initiatives, Osaka Univ.*, [5]*Dep. of Pharmacol., Grad. Sch. of Dentistry, Osaka Univ.*, [6]*Institute for Transdisciplinary Grad. Degree Programs, Osaka Univ.*)

2SCP-4 顕微鏡ライブイメージングと 1 細胞 RNA-seq を組み合わせた自動化システムの開発とシンギュラリティ生物学への応用
An automated system for combining single-cell RNA-seq with live cell imaging and its applications for Singularity Biology
○小川 泰策[1], 城口 克之[1,2]（[1]理研・BDR, [2]理研・IMS）
Taisaku Ogawa[1], Katsuyuki Shiroguchi[1,2] ([1]*RIKEN BDR*, [2]*RIKEN IMS*)

2SCP-5 (2Pos147) Morphodynamic feature space of migrating cells
Daisuke Imoto[1], Nen Saito[2], **Satoshi Sawai**[1,3] ([1]*Graduate School of Arts and Sciences, University of Tokyo*, [2]*Universal Biology Institute, Graduate School of Science, University of Tokyo*, [3]*Research Center for Complex Systems Biology, University of Tokyo*)

2SCP-6 (2Pos243) 上皮メカノケミカル動態の同定
(2Pos243) System identification of mechano-chemical epithelial sheet dynamics
○浅倉 祥文[1], 近藤 洋平[2], 青木 一洋[2], 本田 直樹[1]（[1]京大・生命科学, [2]基生研・定量生物学）
Yoshifumi Asakura[1], Yohei Kondo[2], Kazuhiro Aoki[2], Naoki Honda[1] ([1]*Grad. Sch. Biostudies, Univ. Kyoto*, [2]*Div. Quantitative Biol. ExCELLS, NIBB.*)

14:10〜16:50　D 会場（4F 天葉）／Room D (4F Tenyo)
2SDP　台湾ー日本　二国間シンポジウム：X 線結晶構造解析とクライオ電顕
Taiwan-Japan joint symposium on structural biology using X-ray crystallography and cryo-EM

オーガナイザー：村田 武士（千葉大学），横山 謙（京都産業大学）
Organizers: Takeshi Murata (Chiba University), Ken Yokoyama (Kyoto Sangyo University)

Cryo-electron microscopy (cryo-EM) is rapidly becoming the main technology for studying 3D structures of proteins, while X-ray crystallography is a powerful traditional tool in structural biology. At the same time, we are facing several issues for cryo-EM such as machine-time limitation, and grid preparation difficulties. In this joint symposium, cutting-edge results using these methods are presented by researches from Taiwan and Japan, and we discuss the mutual methodological problems and future view in this field.

はじめに
Opening Remarks

2SDP-1 クライオ電子顕微鏡による繊毛と微小管モーターの解析
Cryo-EM analysis of cilia and microtubule-based motor proteins
○吉川 雅英（東京大学）
Masahide Kikkawa (*The Univ. of Tokyo*)

2SDP-2 Cryo-EM Analysis of a Feline Coronavirus Spike Protein Reveals a Unique Structure and Camouflaging Glycans
Tzu-Jing Yang[1,2], Yen-Chen Chang[1,3], Tzu-Ping Ko[1], Piotr Draczkowski[1], Yu-Chun Chien[1,2], Yuan-Chih Chang[4], Kuen-Phon Wu[1], Kay-Hooi Khoo[1,2], Hui-Wen Chang[3], **Shang-Te Danny Hsu**[1,2] ([1]*Institute of Biological Chemistry, Academia Sinica*, [2]*Institute of Biochemical Sciences, National Taiwan University*, [3]*School of Veterinary Medicine, National Taiwan University*, [4]*Institute of Cellular and Organismic Biology, Academia Sinica*)

2SDP-3 CryoEM studies of bacterial glutamine synthetase
Kuen-Phon Wu, Chia-Wei Chou (*Institute of Biological Chemistry, Academia Sinica*)

2SDP-4 V 型 ATP 合成酵素の膜内在性ドメイン V_o の単粒子解析
Single particle analysis of membrane embedded domain V_o of V-type ATP synthase
○岸川 淳一 [1], 加藤 貴之 [2], 古田 綾 [1], 中西 温子 [1], 光岡 薫 [3], 横山 謙 [1]（[1]京都産業大学 総合生命科学部 生命システム学科, [2]大阪大学大学院 生命機能研究科, [3]大阪大学 超高圧電子顕微鏡センター）
Jun-ichi Kishikawa[1], Takayuki Kato[2], Aya Furuta[1], Atsuko Nakanishi[1], Kaoru Mitsuoka[3], Ken Yokoyama[1] ([1]*Dept. Mol. Biosci., Kyoto Sangyo Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Res. Ctr. UHVEM., Osaka Univ.*)

2SDP-5 胃プロトンポンプの輸送機構に対する構造基盤
Structural basis for the transport mechanism of the gastric proton pump
○阿部 一啓 [1,2]（[1]名古屋大学 細胞生理学研究センター, [2]名古屋大学大学院創薬科学研究科）
Kazuhiro Abe[1,2] ([1]*Cellular and Structural Physiology Institute, Nagoya Univ.*, [2]*Grad. Sch. Pharm. Sci., Nagoya Univ.*)

2SDP-6 PAD4 regulates p53 function through protein citrullination
Chien-Yun Lee[1], Guang-Yaw Liu[2], **Hui-Chih Hung**[1] ([1]*National Chung-Hsing University*, [2]*Chung Shan Medical University*)

おわりに
Closing Remarks

14:10〜16:50　E 会場（4F クリスタルルーム）／Room E (4F Crystal Room)
2SEP　共催：JST さきがけ「量子技術を適用した生命科学基盤の創出」
量子科学で捉える生命現象
Understanding biological systems with quantum science and technology

オーガナイザー：市村 垂生（大阪大学），塗谷 睦生（慶應義塾大学）
Organizers: Taro Ichimura (Osaka University), Mutsuo Nuriya (Keio University)

Recent years have witnessed remarkable progresses in quantum technologies based on quantum science, and these technologies and viewpoints are expected to bring innovations to measurement technologies and interpretations of biological phenomena. However, its application to life science is still in its early days and further leaps are expected in the future. In this symposium, researchers in the field of quantum biology aiming at such attempts will introduce their latest research achievements, and explore the future prospects of the fusion of quantum science and biophysics through discussions with the audiences.

はじめに
Opening Remarks

2SEP-1　Materials chemistry of photo-excited triplet state for dynamic nuclear polarization
Nobuhiro Yanai[1,2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*JST-PRESTO*)

2SEP-2　**溶媒和の量子化学効果の分子動力学シミュレーションの開発と生体分子への応用**
Incorporation of qunatum chemical effect of solvation into molecular dynamics simulation and the applications to biomolecules
○渡邉 宙志 [1,2]（[1]慶應大, [2]JST さきがけ）
Hiroshi Watanabe[1,2] ([1]*Keio Univ. KQCC*, [2]*PRESTO JST*)

2SEP-3　**多光子顕微鏡の応用による脳組織内の分子動態の可視化解析**
Imaging dynamics of molecules inside the brain tissue by the application of multiphoton microscopy
○塗谷 睦生 [1,2,3]（[1]慶應義塾大学医学部, [2]JST さきがけ, [3]横浜国立大学）
Mutsuo Nuriya[1,2,3] ([1]*Keio University School of Medicine*, [2]*JST PRESTO*, [3]*Yokohama National University*)

2SEP-4　Contextuality and Non-Locality in Quantum Physics and Cognitive Science
Yoshihiro Maruyama (*Kyoto University*)

2SEP-5　Label-free molecular vibrational spectro-microscopy
Takuro Ideguchi[1] ([1]*The University of Tokyo*, [2]*PRESTO, JST*)

2SEP-6　Nanoscale thermometry and magnetometry in biology using NV center in diamond
Hitoshi Ishiwata[1,2] ([1]*PRESTO*, [2]*Tokyo Institute of Technology*)

2SEP-7　(2Pos288) **グラフェン電界効果トランジスタとフェムトリットルチャンバーを用いたデバイ遮蔽を超える電気的バイオセンシング**
(2Pos288) Electrical Biosensing beyond the Debye Screening Length Using Graphene Field-Effect Transistor in Femtoliter Microchamber
○小野 堯生 [1], 金井 康 [1], 井上 恒一 [1], 渡邊 洋平 [2], 中北 愼一 [3], 河原 敏男 [4], 鈴木 康夫 [4], 松本 和彦 [1]（[1]阪大産研, [2]京府医大, [3]香川大, [4]中部大）
Takao Ono[1], Yasushi Kanai[1], Koichi Inoue[1], Yohei Watanabe[2], Shin-ichi Nakakita[3], Toshio Kawahara[4], Yasuo Suzuki[4], Kazuhiko Matsumoto[1] ([1]*ISIR, Osaka Univ.*, [2]*Kyoto Pref. Univ of Med.*, [3]*Kagawa Univ.*, [4]*Chubu Univ.*)

14:10〜16:50　F 会場（4F マーブルルーム）／Room F (4F Marble Room)
2SFP　共催：新学術領域研究「進化の制約と方向性」
構成的アプローチを用いた進化研究：拘束と進化可能性の理解へ向けて
Constructive Approaches for Evolution: Toward Understanding of Directionarity and Constraints

オーガナイザー：古澤 力（理化学研究所 / 東京大学），入江 直樹（東京大学）
Organizers: Chikara Furusawa (RIKEN / The University of Tokyo), Naoki Irie (The University of Tokyo)

Living organisms do not evolve in perfectly random directions, instead, we recognize unevenness and directionalities in phenotypic variations and evolutionary changes. However, mechanisms for these directionality or evolutionary constraints remains unclear so far. In this symposium, we will show recent development in this field, in particular, analysis of evolutionary dynamics by constructive approaches to unveil constraints and directionalities in evolution, and discuss current subjects and future perspectives.

2SFP-1　**共生進化生物学の最前線**
Frontiers in experimental evolutionary biology of symbiosis
○深津 武馬（産業技術総合研究所 生物プロセス研究部門）
Takema Fukatsu (*AIST*)

2SFP-2　What makes animal embryos to follow the hourglass model?
Naoki Irie[1,2], Yui Uchida[1,2], Masahiro Uesaka[3] ([1]*Univ. Tokyo, Sch. of Science*, [2]*Univ. Tokyo, Universal Biology Institute*, [3]*RIKEN*)

2SFP-3　Impact of polyploidy on the evolutionary rate
Ryudo Ohbayashi[1], Tetsuhiro Hatakeyama[2] ([1]*BDR., RIKEN*, [2]*Dept. of Basic Sci., Univ. of Tokyo*)

2SFP-4　Analysis of Evolutionary Constraints and Plasticity by Microbial Laboratory Evolution
Chikara Furusawa[1,2] ([1]*BDR, RIKEN*, [2]*UBI, Univ. Tokyo*)

14:10〜16:50　G 会場（4F アイボリールーム）／Room G (4F Ivory Room)
2SGP　ポスト「京」始動を見据えた計算創薬の新展開
New horizon of in-silico drug discovery toward launching post-K computer

オーガナイザー：荒木 望嗣（京都大学），池口 満徳（横浜市立大学）
Organizers: Mitsugu Araki (Kyoto University), Mitsunori Ikeguchi (Yokohama City University)

The drug development process is about to be innovated by launching the post-K supercomputer, which is designed to be the successor of the K computer. In recent years, various next-generation in-silico drug discovery techniques have been developed by combining fundamental molecular-simulation techniques with advanced experimental technologies or artificial intelligences (AIs). In this symposium, the forefront of the in-silico drug discovery will be discussed with young researchers in "Priority issue 1 on Post-K computer" (Building Innovative Drug Discovery Infrastructure Through Functional Control of Biomolecular Systems).

2SGP-1　**拡張アンサンブル法を用いたタンパク質-リガンド結合ポーズの自由エネルギー解析**
Free-energy analysis of protein-ligand binding pose using generalized ensemble methods
○尾嶋 拓, 李 秀栄, 杉田 有治（理研 BDR）
Hiraku Oshima, Suyong Re, Yuji Sugita (*RIKEN BDR*)

2SGP-2 An efficient screening, an accurate evaluation, and a simple prediction of protein complex structures
Kazuhiro Takemura, Akio Kitao (*Sch. Life Sci. Tech., Tokyo Tech.*)

2SGP-3 (2Pos029) Determination of protonated states for native and mutant structures of HIV-1 protease with indinavir by free energy calculations
Masahiko Taguchi, Ryo Oyama, Masahiro Kaneso, Shigehiko Hayashi (*Kyoto University*)

2SGP-4 **創薬標的タンパク質の溶液構造解析**
Ligand-bound forms of drug-discovery target protein in solution studied by molecular dynamics simulations
○浴本 亨[1], 工藤 崇文[1], 山根 努[1], 池口 満徳[1,2]（[1]横浜市大・生命医, [2]理研）
Toru Ekimoto[1], Takafumi Kudo[1], Tsutomu Yamane[1], Mitsunori Ikeguchi[1,2] ([1]*Yokohama City Univ.*, [2]*Riken*)

2SGP-5 **アラニン置換による抗体親和性の向上のメカニズム**
Mechanism of antibody-affinity enhancement through alanine-substitution
○山下 雄史（東京大学）
Takefumi Yamashita (*The University of Tokyo*)

2SGP-6 **タンパク質アポ構造から出発した発展的分子動力学シミュレーションによる薬剤結合モードの予測**
Protein-drug binding mode prediction from the apo-protein structure using a molecular dynamics-based pocket generation approach
○荒木 望嗣[1,2], 奥野 恭史[1,2]（[1]京大・院医, [2]理研・計算科学研究機構）
Mitsugu Araki[1,2], Yasushi Okuno[1,2] ([1]*Grad. Sch. of Med., Kyoto Univ.*, [2]*RIKEN, AICS*)

2SGP-7 Reinforcement Learning and Global Optimization Techniques in Molecular Dynamics Simulations
Kei Terayama[1,2,3], Yasushi Okuno[3], Koji Tsuda[1,4,5] ([1]*AIP, RIKEN*, [2]*MIH, RIKEN*, [3]*Grad. Sch. Med., Kyoto Univ.*, [4]*Grad. Sch. Frontier Sci., Univ. Tokyo*, [5]*NIMS*)

2SGP-8 (2Pos075) **天然変性タンパク質 p53 を標的としたペプチドの人工設計ー液液相分離の制御ー**
(2Pos075) Rational design of peptide targeting intrinsically disordered protein p53 -regulation of function and phase-phase separation-
○鎌形 清人[1], 間野 絵梨子[1], 伊藤 優志[1], 上林 さおり[1], 本多 優也[1], 北原 亮[2], 亀田 倫史[3]（[1]東北大・多元研, [2]立命大・薬, [3]産総研・創薬基盤）
Kiyoto Kamagata[1], Eriko Mano[1], Yuji Itoh[1], Saori Kanbayashi[1], Masaya Honda[1], Ryo Kitahara[2], Tomoshi Kameda[3] ([1]*IMRAM, Tohoku Univ.*, [2]*Coll. Pharmacy Sci., Ritsumeikan Univ.*, [3]*AIRC, AIST*)

14:10〜16:50　H 会場（4F アンバールーム）／Room H (4F Amber Room)

2SHP　可視化デバイス開発と数理モデル化を用いた細胞内アーキテクチャの解読
Decoding intracellular architecture using visualizing device development and mathematical modeling

オーガナイザー：北村 朗（北海道大学），樺山 一哉（大阪大学）
Organizers: Akira Kitamura (Hokkaido University), Kazuya Kabayama (Osaka University)

Cells carry highly organized architectures. Here we introduce the frontier research findings determining the actual image of intracellular architecture such as structural change of chromatin and nucleic acids, enzymatic activity, and signal transduction process via posttranslational modification by visualization using optical imaging and microscopic control devices, or mathematical modeling. We will select more than two subjects from those for the poster presentation (female and/or young researchers are more acceptable), and also we have a comprehensive discussion.

2SHP-1　Deciphering genome organization and dynamics by mathematical modeling and simulation
Soya Shinkai (*RIKEN BDR*)

2SHP-2　Reading out G-quadruplex RNA structure using transient state (TRAST) of photochemical reaction of fluorophores
Akira Kitamura[1,2] ([1]*Lab. Mol. Cell Dynamics, Fac. Adv. Life Sci., Hokkaido Univ.*, [2]*JSPS Scientist for Joint International Research*)

2SHP-3　Isolation and analysis of specific cells, organelles and supramolecular complexes using microfluidic microdroplets
Ryo Iizuka (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

2SHP-4　(2Pos268) 大気圧走査電子顕微鏡 ASEM による骨組織再構築の水中免疫電顕法と cryo-TEM 観察
(2Pos268) Observation of unstained bone tissues and immuno-EM in liquid by ASEM and cryo-TEM
○佐藤 主税 [1], 杉本 真也 [2], 簇野 悠里 [1], 佐藤 真理 [1], 坂井 詠子 [3] ([1]産総研 バイオメディカル, [2]慈恵医大 細菌学, [3]長崎大 歯科薬理学)
Chikara Sato[1], Shinya Sugimoto[2], Yuri Hatano[1], Mari Sato[1], Eiko Sakai[3] ([1]*Biomedical Res. Inst., AIST*, [2]*Dept. Bacteriol., The Jikei Univ. Sch. Med.*, [3]*Dental Pharmacology, Nagasaki Univ.*)

2SHP-5　(2Pos246) 細胞内のインスリン様成長因子-I(IGF-I)シグナルは振動する
(2Pos246) Cellular insulin-like growth factor-I (IGF-I) signal can be oscillated
○増田 正人, 伯野 史彦, 高橋 伸一郎（東大・院農生科・応動）
Masato Masuda, Fumihiko Hakuno, Shin-Ichiro Takahashi (*Dep. App. Ani. Sci., Grad. Sch. Agr. Lif. Sci., The Univ. Tokyo*)

2SHP-6　入力制御によるライブセルイメージング解析
Live cell imaging analyses by input control system
○樺山 一哉 [1,2,3]（[1]大阪大学大学院 理学研究科 化学専攻, [2]大阪大学大学院 理学研究科 基礎理学プロジェクト研究センター, [3]大阪大学 放射線科学基盤機構）
Kazuya Kabayama[1,2,3] ([1]*Department of Chemistry, Graduate School of Science, Osaka University*, [2]*Project Research Center, Graduate School of Science, Osaka University*, [3]*Institute for Radiation Sciences, Osaka Univeresity*)

おわりに
Closing Remarks
北村 朗（北大）
Akira Kitamura (*Hokkaido Univ.*)

3 日目（9 月 26 日（木））／Day 3（Sep. 26 Thu.）

8:30〜11:10　B 会場（4F 天玉）／Room B (4F Tengyoku)
3SBA　**ヘム蛋白質の機能を司る構造・ダイナミクスとエネルギー流：理論と実験**
Structure, Dynamics and Energy Flow that Govern Heme Protein Functions: Theory and Experiments

オーガナイザー：倭 剛久（名古屋大学），David Leitner（University of Nevada）
Organizers: Takahisa Yamato (Nagoya University), David Leitner (University of Nevada)

Within a thermally fluctuating protein molecule under physiological conditions, tightly packed amino acid residues are interacting with each other exchanging energies between them. Thanks to the recent developments in theoretical/computational/experimental techniques, biophysical mechanisms of protein functions have been elucidated at atomic detail. In particular, heme proteins provide an ideal research targets for biophysicists because of their natural “probe” build in a protein matrix. In this symposium, we would like to discuss recent advancement of biophysical studies on heme proteins and molecular basis of their functions.

3SBA-1　Theoretical model of the allosteric transition of oxygen sensor domain of FixL
Takahisa Yamato (*Nagoya University*)

3SBA-2　Molecular Mechanism of NO Reduction by Nitric Oxide Reductase in Cellular System
Yoshitsugu Shiro (*Univ. of Hyogo*)

3SBA-3　Direct observation of vibrational energy flow in hemeproteins
Misao Mizuno (*Grad. Sch. Sci., Osaka Univ.*)

3SBA-4　**ミオグロビンとヘモグロビンの結晶内光解離反応過程の観測**
Observation of photolysis reaction of myoglobin and hemoglobin in crystals
○佐藤 文菜（自治医大）
Ayana Sato-Tomita (*Jichi Med. Univ.*)

3SBA-5　Watching energy transport in proteins: Identifying dynamic networks and thermodynamic properties
David Leitner (*University of Nevada, Reno*)

8:30〜11:10　C 会場（4F 天樹）／Room C (4F Tenjyu)

3SCA　生体運動の多様性と普遍性―細胞内ダイナミクスから集団運動まで―
Diversity and universality of motile mechanism of living things: From intracellular dynamics to collective motion

オーガナイザー：中村 修一（東北大学），鹿毛 あずさ（豊橋技術科学大学）
Organizers: Shuichi Nakamura (Tohoku University), Azusa Kage (Toyohashi University of Technology)

'Why living things move?': this question has attracted many people since the ancient Greek times. In addition to major types of cell motility such as swimming and crawling, recent studies have revealed that many organisms adopt unique mechanisms of cell motility. Moreover, reconstitution approach and mathematical modelling aim to reproduce cell-like movements. In this session, we would like to discuss diversity and universality of motile mechanism of living things in reference to such diverse studies.

はじめに
Opening Remarks
鹿毛 あずさ（豊橋技術科学大学）
Azusa Kage (*Toyohashi Univ. of Tech.*)

3SCA-1　君子は豹変す: シアノバクテリアも心変わりする
Cyanobacteria change their mind
○中根 大介, 西坂 崇之（学習院大・理・物理）
Daisuke Nakane, Takayuki Nishizaka (*Dept. Phys., Gakushuin Univ.*)

3SCA-2　Visualization of bacteria motility strategies and biofilm formation in tight microfluidic environments
Andrew Utada (*Univ. of Tsukuba*)

3SCA-3　アーキアべん毛の生物物理学的視点による特性評価
Biophysical characterization of molecular motors in archaea
○木下 佳昭 [1,2], Helen Miller[1], Zhengqun Li[2], 三上 渚 [2], Quax E.F. Tessa[2], Albers Sonja-Verena[2], Berry Richard[1]（[1]オックスフォード大学, [2]フライブルク大学）
Yoshiaki Kinosita[1,2], Miller Helen[1], Li Zhengqun[2], Nagisa Mikami[2], E.F. Tessa Quax[2], Sonja-Verena Albers[2], Richard Berry[1] ([1]*Oxford University*, [2]*University of Freiburg*)

3SCA-4　バクテリア細胞質のガラス的動力学の代謝活動による流動化現象について
Glassy dynamics of a model of bacterial cytoplasm with metabolic activities
○大山 倫弘 [1], 川崎 猛史 [2], 水野 英如 [3], 池田 昌司 [3]（[1]産業技術総合研究所, [2]名古屋大学, [3]東京大学）
Norihiro Oyama[1], Takeshi Kawasaki[2], Hideyuki Mizuno[3], Atsushi Ikeda[3] ([1]*AIST*, [2]*Nagoya University*, [3]*University of Tokyo*)

3SCA-5　ボルボックス目緑藻の光行動
Photomovements of *Chlamydomonas*, *Volvox* and *Tetrabaena*
○若林 憲一（東工大・化生研）
Ken-ichi Wakabayashi (*CLS, Tokyo Tech*)

3SCA-6　Collective swimming of living spinners
Azusa Kage[1], Takayuki Torisawa[2,3], Ayano A. Medo[4,5], Ken H. Nagai[6] ([1]*TUT*, [2]*NIG*, [3]*SOKENDAI*, [4]*U Hyogo*, [5]*Present address: Kyoto U*, [6]*JAIST*)

3SCA-7　魚類表皮の遊走細胞ケラトサイトのストレスファイバ車輪の回転
Rotation of stress fiber-wheel in migrating fish keratocytes
○沖村 千夏（山口大・理）
Chika Okimura (*Fac. Sci., Yamaguchi Univ.*)

8:30〜11:10　D 会場（4F 天葉）／Room D (4F Tenyo)
3SDA　生命現象の理解を目指す光遺伝学の新展開
Optogenetics: Applying photoreceptor for understanding biological phenomena

オーガナイザー：角田 聡（名古屋工業大学），井上 圭一（東京大学）
Organizers: Satoshi Tsunoda (Nagoya Institute of Technology), Keiichi Inoue (The University of Tokyo)

This symposium is aimed to introduce the cutting edge technology in optogenetics. Optogenetics markedly revolutionized life science. This technique allows fast and precise control of defined biological event, such as neuronal excitation, cell locomotion and gene expression, even in complex system such as freely moving animals. Optogenetics has been realized through understanding molecular properties of photoreceptors, developing new optical technique, genetics in model systems and modern brain science. In this symposium, we gather scientist with diverse expertise to discuss existing and newly emerging approaches which open new landscape for study of biology in future.

3SDA-1　オルガネラ・オプトジェネティクス - 細胞内 Ca^{2+} ダイナミクスの光操作
Organelle optogenetics: Optical manipulation of intracellular Ca^{2+} dynamics
○八尾 寛 [1], 浅野 豪文 [2], 五十嵐 敬幸 [3], 石塚 徹 [4]（[1]東大・物性研, [2]東京医歯大・細胞生物, [3]ウェスタン大・シューリッチ医歯校, [4]東北大・生命）
Hiromu Yawo[1], Toshifumi Asano[2], Hiroyuki Igarashi[3], Toru Ishizuka[4] ([1]*ISSP, Univ. Tokyo*, [2]*Cell Biol., TMDU*, [3]*Western Univ., Schulich Sch. Med. Dent., Canada*, [4]*Tohoku Univ. Grad. Sch. Life Sci.*)

3SDA-2　光遺伝学ツールとしての光サイクル型動物オプシンの最適化
Engineering of photocyclic animal opsin as a potential optogenetic tool
○山下 高廣（京大・院理）
Takahiro Yamashita (*Grad. Sch. of Sci., Kyoto Univ.*)

3SDA-3　(3Pos142) 生体組織への応用が期待される光感度の高いチャネルロドプシン
(3Pos142) Novel optogenetics tool: A light-gated cation channel with high-reactivity to weak light
○細島 頌子 [1], 重村 竣太 [1], 神取 秀樹 [1], 角田 聡 [1,2]（[1]名古屋工業大学, [2]JST, さきがけ）
Shoko Hososhima[1], Shunta Shigemura[1], Hideki Kandori[1], Satoshi Tsunoda[1,2] ([1]*Nagoya Institute of Technology*, [2]*JST, PRESTO*)

3SDA-4　Optical Switches of Membrane Receptor Activities Using CRY2
Takeaki Ozawa (*Dept. Chem., Univ. Tokyo*)

3SDA-5　腹側被蓋野の GABA 作動性神経がノンレム睡眠を調節する
VTA-GABA neurons regulate NREM sleep
○山中 章弘 [1,2]（[1]名古屋大学環境医学研究所, [2]科学技術振興機構 CREST）
Akihiro Yamanaka[1,2] ([1]*RIEM, Nagoya University*, [2]*CREST, JST*)

3SDA-6 (1Pos139) 集団細胞遊走における機械的なシグナルを介した ERK 活性伝播
(1Pos139) ERK activation waves mediated by intercellular mechanical signals during collective cell migration
○日野 直也 [1], Trepat Xavier[2], 松田 道行 [1,3], 平島 剛志 [3]（[1]京大・院生命科学, [2]IBEC, Spain, [3]京大・院医学）
Naoya Hino[1], Xavier Trepat[2], Michiyuki Matsuda[1,3], Tsuyoshi Hirashima[3] ([1]*Grad. Sch. of Biostudies, Kyoto Univ.*, [2]*IBEC, Spain*, [3]*Grad. Sch. of Med., Kyoto Univ.*)

8:30〜11:10　E 会場（4F クリスタルルーム）／Room E (4F Crystal Room)
3SEA　共催：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（温度生物学）」
温度を基軸とした生物物理現象の理解
Thermal Biology

オーガナイザー：原田 慶恵（大阪大学），岡部 弘基（東京大学）
Organizers: Yoshie Harada (Osaka University), Kohki Okabe (The University of Tokyo)

Temperature has attracted great attention in the search for deeper understanding of various life activities. In recent years, the emergence of thermometric methodologies for and insights into the thermal response of cellular organelles has opened a door for thermal biology at the single cell level. In this symposium, through the introduction of the challenges in single-cell thermal biology faces when exploring the mechanisms of thermal sensation and response inside a cell, we will discuss the fundamental principles of how temperature facilitates cell functions.

3SEA-1 単一細胞内温度シグナリングによるストレス顆粒形成の分子機構
The molecular mechanism of thermal signaling-dependent SG formation in single cells
○岡部 弘基 [1,2]（[1]東京大学大学院薬学系研究科, [2]JST さきがけ）
Kohki Okabe[1,2] ([1]*Grad. Sch. Pharma. Sci., The Univ. of Tokyo*, [2]*PRESTO, JST*)

3SEA-2 (1Pos263) ラマンイメージングを用いた細胞内の水の可視化とラベルフリー細胞内温度測定への応用
(1Pos263) Raman imaging of water in a cell and its application to label-free evaluation of intracellular temperature
○杉村 俊紀, 梶本 真司, 中林 孝和（東北大院・薬）
Toshiki Sugimura, Shinji Kajimoto, Takakazu Nakabayashi (*Grad. Sch. Pham. Sci., Tohoku. Univ*)

3SEA-3 (3Pos064) 温度上昇とテラヘルツ光照射は転写反応に異なる影響を及ぼす。
(3Pos064) Terahertz radiation and temperature increase differently affect transcription by RNA polymerase
○今清水 正彦 [1], 田中 真人 [1], 保科 宏道 [2], 竹内 恒 [1]（[1]産総研, [2]理研）
Masahiko Imashimizu[1], Masahito Tanaka[1], Hiromichi Hoshina[2], Koh Takeuchi[1] ([1]*AIST*, [2]*RIKEN*)

3SEA-4 小胞体-ミトコンドリア間クロストークを介した褐色脂肪細胞の機能制御
Regulation of brown adipocyte function through the crosstalk signaling between mitochondria and the endoplasmic reticulum
○西頭 英起（宮崎大・医・機能生化）
Hideki Nishitoh (*Lab. of Biochem. and Mol. Biol., Dept. of Med. Sci., Univ. of Miyazaki*)

3SEA-5 膜脂質を介する細胞内温度の制御機構
Membrane lipid-mediated regulation of intracellular temperature
村上 光, 長尾 耕治郎, ○梅田 眞郷（京都大学）
Akira Murakami, Kohjiro Nagao, **Masato Umeda** (*Kyoto University*)

おわりに
Closing Remarks

8:30〜11:10　F 会場（4F マーブルルーム）／Room F (4F Marble Room)
3SFA　ナノ空間の生物物理
Biophysics in Nano-space

オーガナイザー：多田隈 尚史（大阪大学），北川 大樹（東京大学）
Organizers: Hisashi Tadakuma (Osaka Univ.), Daiju Kitagawa (The University of Tokyo)

Biomolecules such as proteins and nucleic acids are nano-meter sized materials. Therefore, it is important to understand how these biomolecules interact with each other and function in a specific environment. Recently, cutting-edge technologies and new approaches open the frontier of biophysics. Here, we will discuss the novel factors and phenomena in nano-space driving the biological activities.

3SFA-1　How nano-space affects biological phenomea
Hisashi Tadakuma (*IPR, Osaka University*)

3SFA-2　グアニン四重鎖と i-モチーフ構造を分子プローブとして使ったナノ空間の物性の検討
Investigation of physical properties of a confined nanospace using G-quadruplex and i-motif as a molecular probe
○遠藤 政幸（京大・院理）
Masayuki Endo (*Grad. Sch. Sci. Kyoto Univ.*)

3SFA-3　A widespread family of heat-resistant obscure (Hero) proteins protect against protein instability and aggregation
Kotaro Tsuboyama[1], Shintaro Iwasaki[2], Yukihide Tomari[1] ([1]*UTokyo IQB RNA function lab*, [2]*Riken RNA systems biochemical lab*)

3SFA-4　有限体積下で働く分子システム設計：人工細胞モデル構築を通して
Molecular system design that works under finite volume: through artificial cell model construction
○野村 慎一郎（東北大学大学院 工学研究科 ロボティクス専攻）
Shin-ichiro Nomura (*Dep. Robotics, TOHOKU Univ.*)

3SFA-5　(3Pos195) Intracellular delivery of biologics using magnetically-navigated nanocarrier
Yoshihiro Sasaki, Ryosuke Mizuta, Naoya Kinoshita, Kazunari Akiyoshi (*Department of Polymer Chemistry, Graduate School of Engineering, Kyoto University*)

3SFA-6　Using SABER to amplify multiplexed FISH signal from RNA and DNA targets
Jocelyn Y Kishi[1,2,5], Sylvain W. Lapan[3,5], Brian J. Beliveau[1,2,5], Emma R. West[3,5], Allen Zhu[1,2], Hiroshi M. Sasaki[1,2], Sinem K. Saka[1,2], Yu Wang[1,2], Constance L. Cepko[3,4], Peng Yin[1,2] ([1]*Wyss Institute, Harvard Univ.*, [2]*Dept. Systems Biology, Harvard Medical School*, [3]*Dept. Genetics, Blavatnik Institute, Harvard Medical School*, [4]*Howard Hughes Medical Institute*, [5]*These authors contributed equally*)

3SFA-7 (3Pos075) 三次元構造モデルから発生過程における細胞機能の理解を試みる
(3Pos075) Attempt to understand the cellular function during developmental process from 3D structural model
黒田 純平[1,4], 板橋 岳志[1,2,3], 一ノ瀬 孝子[1], 近藤 滋[4], ○岩根 敦子[1,2,3]（[1]理研・BDR・細胞場, [2]広大院・統合生命科学, [3]阪大院・生命機能, [4]阪大院・生命機能・パターン形成）
Junpei Kuroda[1,4], Takeshi Itabashi[1,2,3], Takako M. Ichinose[1], Shigeru Kondo[4], **Atsuko H. Iwane**[1,2,3] ([1]*Cell Field Struc., BDR, Riken*, [2]*Grad. sch. Integ. Sci. Life, Hiroshima Univ.*, [3]*Spec. Res. Promot. Group, Grad. Sch. Fronti., Biosci., Osaka Univ.*, [4]*Pattern formation, Grad. Sch. Fronti., Biosci., Osaka Univ.*)

3SFA-8 ナノスケール空間における中心小体複製メカニズム
Mechanisms of centriole duplication in nano-space
○北川 大樹, 山本 昌平, 高尾 大輔（東京大学大学院薬学系研究科 生理化学教室）
Daiju Kitagawa, Shohei Yamamoto, Daisuke Takao (*Graduate School of Pharmaceutical Sciences, The University of Tokyo*)

8:30〜11:10　G 会場（4F アイボリールーム）／Room G (4F Ivory Room)
3SGA　超解像顕微鏡による生物物理学的生理学・病理学
Biophysical Physiology and Pathology by the Application of Superresolution Microscopy

オーガナイザー：角山 貴昭（沖縄科学技術大学院大学），笠井 倫志（京都大学）
Organizers: Taka A. Tsunoyama (OIST), Rinshi S. Kasai (Kyoto University)

Superresolution microscopy is widely used now a days, while its contributions for biophysics are still smaller than single molecule imaging. However, superresolution microscopy has the potential for solving the problems in physiology and pathology from biophysical point of view. In this symposium, the speakers are leading researchers in the field, and we expect innovative ideas and fruitful discussions.

3SGA-1 Advancing molecular medicine with quantitative single molecule localization microscopy
Devin L. Wakefield[1], Kathleen M. Lennon[1], Steven J. Tobin[1], Matthew S. Brehove[1], Adam L. Maddox[1], Ajay Goel[3], Kendall Van Keuren-Jensen[2], Daniel Schmolze[1], **Tijana Jovanovic-Talisman**[1] ([1]*City of Hope*, [2]*TGen*, [3]*Baylor Research Institute*)

3SGA-2 High resolution systems approach to discover mitotic regulation of the nucleus
Paul S. Maddox (*Department of Biology, University of North Carolina at Chapel Hill*)

3SGA-3 Actin-induced compartments and islands in focal adhesions as revealed by simultaneous ultrafast PALM and single-molecule tracking
Takahiro Fujiwara (*WPI-iCeMS, Kyoto Univ.*)

3SGA-4 The axonal cytoskeleton at the nanoscale
Christophe Leterrier (*INP CNRS-AMU UMR7051*)

3SGA-5 (3Pos179) 転写伸長を制御するメディエーターの 1 分子超解像イメージングによる分子局在と動態の定量解析
(3Pos179) Molecular localization and dynamics of Mediator regulating transcription elongation using single-molecule and super-resolution microscopy
伊藤 由馬[1], 國見 慎之介[1], 高橋 秀尚[2], ○徳永 万喜洋[1]（[1]東工大・生命理工学院, [2]横浜市大・院医学）
Yuma Ito[1], Shinnosuke Kunimi[1], Hidehisa Takahashi[2], **Makio Tokunaga**[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*)

8:30〜11:10　H 会場（4F アンバールーム）／Room H (4F Amber Room)
3SHA　タンパク質の“質”を評価する
The Quality of Proteins - Multiple Approaches for Protein Evaluation -

オーガナイザー：小川 覚之（東京大学），内橋 貴之（名古屋大学）
Organizers: Tadayuki Ogawa (The University of Tokyo), Takayuki Uchihashi (Nagoya University)

Proteins undergo a variety of their qualitative changes throughout their life. The variety of their “quality” provide their specific function and behavior, which forms the basis of biological systems. Therefore, accurate and precise analyses of the quality of proteins will deepen our knowledge in the fundamental behavior of the protein molecules in biological systems and pathogenesis. From this perspective, this session focuses on the multiple protein analyses on the protein quality, including supra-structure in solution, post-translational modification, pathogenic stress, aging, etc., from the basic research to the medical and industrial applications, and discuss about the comprehensive usage of multiple analyses.

はじめに
Opening Remarks
小川 覚之（東京大）
Tadayuki Ogawa (*Univ. of Tokyo*)

3SHA-1　ヨウ素染色によるアミロイド線維構造多形と構造伝播の解析の試み
Iodine staining as a useful probe for amyloid polymorphism and its propagation
○茶谷 絵理 [1], 平松 貴人 [1], 柚 佳祐 [1], 山本 直樹 [2]（[1]神戸大院理, [2]自治医大医）
Eri Chatani[1], Takato Hiramatsu[1], Keisuke Yuzu[1], Naoki Yamamoto[2] ([1]*Grad. Sch. Sci., Kobe Univ.*, [2]*Fac. Med., Jichi Medical Univ.*)

3SHA-2　フィブロインタンパク質からクモ糸への人工的再構成
Artificial reconstitution of the multi-hierarchical structure in spider silk from fibroin proteins
○上久保 裕生 [1,2], 佐藤 健大 [3]（[1]奈良先端大 物質創成, [2]高エネ機構 物構研, [3]Spiber 株式会社）
Hironari Kamikubo[1,2], Takehiro Sato[3] ([1]*NAIST MS*, [2]*KEK IMSS*, [3]*Spiber Inc.*)

3SHA-3　Biophysical characterization of environment-dependent protein assemblies of physiological and pathological interest
Maho Yagi-Utsumi, Koichi Kato (*ExCELLS, NINS*)

3SHA-4　(3Pos096) 過渡的に形成される GPCR ダイマーの研究：細胞内蛍光 1 分子観察によるアプローチ
(3Pos096) Examining the transiently formed GPCR dimer: an approach by single fluorescent molecule observation in living cells
○笠井 倫志（京大 ウイ・再生研）
Rinshi Kasai (*Inst. Front. Life. Med. Sci., Kyoto Univ.*)

3SHA-5　(3Pos014) STAP-2 により Breast tumor kinase が活性化する機構の解明
(3Pos014) Molecular basis of Breast tumor kinase by an adaptor protein, STAP-2
○中迫 純希 [1], 松尾 友樹 [2], 神田 諒 [2], 田中 睦乃 [2], 姚 閔 [3], 松田 正 [2], 前仲 勝実 [2], 尾瀬 農之 [2,3,4]（[1]北大院 生命科学, [2]北大院 薬, [3]北大院 先端生命, [4]JST さきがけ）
Junki Nakasako[1], Yuki Matsuo[2], Ryo Kanda[2], Yoshino Tanaka[2], Min Yao[3], Tadashi Matsuda[2], Katsumi Maenaka[2], Toyoyuki Ose[2,3,4] ([1]*Graduate school of Life Science*, [2]*Faculty of Pharm.*, [3]*Faculty of Advanced Life Science, Hokkaido University*, [4]*JST PRESTO*)

3SHA-6　Visualization of Qualitative Change of Proteins with High-Speed Atomic Force Microscopy
Takayuki Uchihashi[1,2] ([1]*Department of Physics*, [2]*ExCELLS, NINS*)

3SHA-7 Aggregation and misfolding of therapeutic antibodies in bioprocessing

Masayoshi Onitsuka[1,2] ([1]*Grad. Sch. of Tech. Ind. Soc. Sci., Tokushima Univ.*, [2]*Manufacturing Technology Association of Biologics*)

ポスター　Poster

1 日目（9 月 24 日（火））／Day 1（Sep. 24 Tue.）
4F 天瑞・ホワイエ／4F TENZUI・Foyer

蛋白質：構造／Protein: Structure

1Pos001* HDX-MS を用いた Fc 断片と IgG1 全長の Fc 領域における構造解析
Structural analysis of IgG Fc region in Fc fragment and IgG1 full-body by HDX-MS
Yuki Yamaguchi[1], Tesuo Torisu[1], Susumu Uchiyama[1,2] ([1]*Grad. Sch., Eng., Univ. Osaka*, [2]*ExCELLS*)

1Pos002* 呼吸鎖における拡張型超複合体の Cryo-EM による構造の解明
Elucidation of the structure of extended super-complex in the respiratory chain by cryo-EM
Kasumi Hirakawa, Wataru Ishibashi, Tomoichirou Kusumoto, Junshi Sakamoto, Takuo Yasunaga (*Grad. Sch Comp. Sci. and Sys. Eng, Kyushu Inst. Tech.*)

1Pos003* シゾロドプシンのプロトン輸送の構造基盤
Structural basis of proton transport in Schizorhodopsin
Akimitsu Higuchi[1], Wataru Shihoya[1], Keiichi Inoue[2,3,4,5], Masae Konno[2], Hideki Kandori[2,3], Osamu Nureki[1] ([1]*Department of Biological Sciences, Graduate School of science, University of Tokyo*, [2]*Department of Life Science and Applied Chemistry, Nagoya Institute of Technology*, [3]*OptoBioTechnology Research Center, Nagoya Institute of Technology*, [4]*The Institute for Solid State Physics, The University of Tokyo*, [5]*PRESTO, Japan Science and Technology Agency*)

1Pos004* カチオン性抗菌ペプチド Hymenochirin-1Pa および変異体 D9K の細菌膜結合構造と膜選択性の解析
Membrane-bound structure and membrane selectivity of cationic antimicrobial peptide Hymenochirin-1Pa and its analog D9K
Akifumi Ohyama[1], Batsaikhan Mijiddorj[2,3], Kazuyoshi Ueda[2], Akira Naito[2], Izuru Kawamura[1,2] ([1]*Grad. Sch. Eng. Sci., Yokohama Natl. Univ*, [2]*Grad. Sch. Eng., Yokohama Natl. Univ*, [3]*Sch. Eng. Appl. Sci., Natl. Univ. Mongolia*)

1Pos005 (1SCP-5) The role of C-terminal carboxylation in α-conotoxin LsIA interactions with human α7 nicotinic acetylcholine receptor *in silico*
Jierong Wen, Andrew Hung (*Sch. Sci., RMIT Univ.*)

1Pos006 分子動力学法を用いた Hras-GTP/GDP 複合体の各部の構造変化と各部の水素結合との同時緩和モードの研究
Molecular dynamics study of simultaneous relaxation modes between structures and the hydrogen bonds in the Hras-GTP/GDP complexes
Takeshi Miyakawa[1], Ryota Morikawa[1], Masako Takasu[1,2], Kimikazu Sugimori[2], Kazutomo Kawaguchi[3], Hidemi Nagao[3] ([1]*Sch. of Life Sci., Tokyo Univ. of Pharm. and Life Sci.*, [2]*Inst. of Liberal Arts. & Sci., Kanazawa Univ.*, [3]*Coll. of Sci. and Eng., Kanazawa Univ.*)

1Pos007 自動デザイン：分子シミュレーションデータを用いた自動ドラッグデザイン
AutoDesign - an automated drug design by using protein-ligand simulation data
Hironori Kokubo, Naoki Miyamoto, Yoshi Nara (*Axcelead, Inc.*)

1Pos008 Simulating large-amplitude transitions in proteins with a coarse-grained model
Ai Shinobu[1], Chigusa Kobayashi[1], Yasuhiro Matsunaga[2], Yuji Sugita[1,3,4] ([1]*RIKEN Center for Computational Science*, [2]*Saitama Univ., Grad. Sch. Sci. Eng.*, [3]*RIKEN Cluster for Pioneering Research*, [4]*RIKEN Center for Biosystems Dynamics Research*)

1Pos009 Dynamics and interdomain interactions in a Drosophila adapter protein (Drk) and their correlation to the unfolding of the N-SH3 domain
Hisham Dokainish[1], Yusuke Suemoto[2], Teppei Ikeya[2], Takuma Kasai[3], Takanori Kigawa[3], Yutaka Ito[2], Yuji Sugita[1] ([1]*Riken, Theoretical Molecular Science Laboratory*, [2]*Department of Chemistry, Tokyo Metropolitan University*, [3]*RIKEN, Center for Biosystems Dynamics Research*)

1Pos010 微小管内タンパク質によるチューブリン格子構造の内側からの制御
Microtubule inner proteins regulate the tubulin lattice architecture from the inside
Muneyoshi Ichikawa[1], Ahmad Khalifa[2], Shintaroh Kubo[3], Kaustuv Basu[2], Daniel Dai[2], Amin Maghrebi[2], Javier Vargas[2], Khanh-Huy Bui[2] ([1]*Dept. of Systems Biol., NAIST*, [2]*McGill Univ.*, [3]*Dept. Biophysics, Kyoto Univ.*)

1Pos011 小角散乱によるアミロイド線維中のヒト α-シヌクレインの構造解析
Structural analysis of human α-synuclein within amyloid fibrils by small-angle scattering
Satoru Fujiwara[1], Tatsuhito Matsuo[1], Yasunobu Sugimoto[2] ([1]*Inst. Quantum Life Science, QST*, [2]*Nagoya Univ.*)

1Pos012 共溶媒の構造類似度を利用した共溶媒分子動力学法における密度マップの類似度の推定
Estimation of the probability map (Pmap) similarity of cosolvent MD (CMD) from structural similarities of cosolvents
Keisuke Yanagisawa[1], Yoshitaka Moriwaki[1], Tohru Terada[2,3], Kentaro Shimizu[1,2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*, [3]*Agr. Bioinfo. Res. Unit., Grad. Sch. Agr. Life Sci., Univ. Tokyo*)

1Pos013 単量体タンパク質の長時間シミュレーションのフォールディングパスウェイの動的解析
Dynamical analysis on the folding pathways of long simulations of a single protein
Ayori Mitsutake[1], Hiroshi Takano[2] ([1]*Meiji Univ.*, [2]*Keio Univ.*)

1Pos014 Targeting the cryptic sites: NMR-based strategy to improve the draggability of proteins by controlling the conformational equilibrium
Koh Takeuchi[1], Yumiko Mizukoshi[2], Yuji Tokunaga[1], Hitomi Matsuo[2], Ichio Shimada[3] ([1]*AIST, molprof*, [2]*JBiC*, [3]*The Univ. Tokyo, Grad Sch Pharm Sci*)

1Pos015 ヘリオロドプシンの構造と生物物理学的解析
Structure and biophysical characterization of the heliorhodopsin
Wataru Shihoya[1], Keiichi Inoue[2,3,4,5], Singh Manish[2], Masae Konno[2], Shoko Hososhima[2], Keitaro Yamashita[1], Kento Ikeda[6], Akimitsu Higuchi[1], Sae Okazaki[1], Izume Tamaki[1], Masanori Hashimoto[2], Ritsu Mizutori[2], Sahoko Tomida[2], Yumeka Yamauchi[2], Rei Abe-Yoshizumi[2], Kota Katayama[2,3], P. Satoshi Tsunoda[2], Mikihiro Shibata[7,8], Yuji Furutani[2,9,10], Alina Pushkarev[11], Oded Beja[11], Takayuki Uchihashi[12,13], Hideki Kandori[2,3], Osamu Nureki[1] ([1]*Department of Biological Sciences, Graduate School of Science, The University of Tokyo*, [2]*Department of Life Science and Applied Chemistry, Nagoya Institute of Technology*, [3]*OptoBioTechnology Research Center, Nagoya Institute of Technology*, [4]*The Institute for Solid State Physics, The University of Tokyo*, [5]*RESTO, Japan Science and Technology Agency*, [6]*School of Mathematical and Physical Sciences, Graduate School of Natural Science & Technology, Kanazawa University*, [7]*Nano Life Science Institute (WPI-NanoLSI), Kanazawa University*, [8]*High-speed AFM for Biological Application Unit, Institute for Frontier Science Initiative, Kanazawa University*, [9]*Department of Life and Coordination-Complex Molecular Science, Institute for Molecular Science, National Institutes of Natural Sciences*, [10]*Department of Structural Molecular Science, The Graduate University for Advanced Studies (SOKENDAI)*, [11]*Israel Institute of Technology*, [12]*Department of Physics, Nagoya University*, [13]*Structural Biology Research Center, Graduate School of Science, Nagoya University*)

1Pos016 クロストリジウム属 2 成分毒素輸送チャネル Ib ポアのクライオ電子顕微鏡構造解析
Cryo-EM structure of clostridial binary toxin translocation channel Ib-pore
Tomohito Yamada[1], Toru Yoshida[1], Akira Kawamoto[2], Kaoru Mitsuoka[3], Kenji Iwasaki[4], Hideaki Tsuge[1] ([1]*Sch. Life Sci., Univ. Kyoto-sangyo*, [2]*Protein inst., Univ. Osaka*, [3]*Research Center for Ultra-High Voltage Electron Microscopy, Univ. Osaka*, [4]*Life Science Center for Survival Dynamics, Tsukuba Advanced Research Alliance, Univ. of Tsukuba*)

1Pos017 藍色細菌 *Anabaena variabilis* 由来 RNA 結合タンパク質の二次構造
Secondary structure of cyanobacterial RNA-binding protein, RbpD, from *Anabaena variabilis*
Hayato Morita[1], Toshihiko Sugiki[2], Chojiro Kojima[2,3], Hidenori Hayashi[4], Naoki Sato[5] ([1]*Fac. Sci., Josai. Univ.*, [2]*Inst. Prot. Res., Osaka Univ.*, [3]*Fac. Eng. Yokohama. Nat. Univ.*, [4]*Grad. Sch. Sci. Eng., Ehime Univ.*, [5]*Grad. Sch. Arts. Sci., Univ. Tokyo*)

1Pos018 Hydrogen bond donors and acceptors are generally depolarized in α-helices as revealed by a negative fragmentation approach
Yu Takano[1,2], Hiroko X. Kondo[1,3], Ayumi Kusaka[2], Shusuke Yamanaka[4], Nakamura Haruki[2] ([1]*Grad. Sch. Info. Sci., Hiroshima City U.*, [2]*IPR, Osaka U.*, [3]*Faculty Eng. Kitami Inst. Tech.*, [4]*Grad. Sch. Sci., Osaka U.*)

1Pos019 Full atomistic model building of EhV-ATPase using homology modeling/molecular dynamics simulation based on the low resolution cryoEM map
Yu Yamamori[1], Jun Tsunoda[2,3], Ray Burton-Smith[3], Chihong Song[3], Ryouta Iino[2,4], Kazuyoshi Murata[2,3], Kentaro Tomii[1] ([1]*AIST*, [2]*SOKENDAI*, [3]*NIPS*, [4]*IMS*)

1Pos020 2 次元テンプレートマッチング法によるクロマチン構造多型解析への挑戦
Challenging for multi-conformational analysis of chromatin using the two-dimensional template matching method
Atushi Tokuhisa[1,2,3], Ryo Kanada[1], Shuntaro Chiba[2], Kei Terayama[3,4,5], Shigeyuki Matsumoto[2], Yuta Isaka[1,6], Biao Ma[1,6], Narutoshi Kamiya[7], Yasushi Okuno[1,3,5,6] ([1]*RCH, RIKEN*, [2]*MIH,RIKEN*, [3]*R-CCS,RIKEN*, [4]*AIP,RIKEN*, [5]*Medicine,Kyoto U*, [6]*CCD,FBRI*, [7]*Simulation,U.Hyogo*)

1Pos021 (1SFP-4) 創薬標的タンパク質の中性子結晶構造解析
(1SFP-4) Neutron crystallographic analysis of drug-target proteins
Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

1Pos022 Crystal structure of human Dishevelled1 PDZ with its inhibitor
Shotaro Yasukochi[1], Nobutaka Numoto[2], Kiminori Hori[1], Natsuko Tenno[1], Takeshi Tenno[1], Nobutoshi Ito[2], Hidekazu Hiroaki[1] ([1]*Grad. Sch. Pharm Sci., Univ. Nagoya*, [2]*Med Res Inst., TMDU*)

蛋白質：構造機能相関／Protein: Structure & Function

1Pos023* 拡張アンサンブル法によるビタミン D 受容体のアゴニスト/アンタゴニスト活性調節機構の研究
Regulation mechanism of agonistic / antagonistic activities of vitamin D receptor studied by generalized ensemble method
Takafumi Kudo[1], Toru Ekimoto[1], Tsutomu Yamane[1], Mitsunori Ikeguchi[1,2] ([1]*Grad. Sch. Med Life Sci., Yokohama City Univ.*, [2]*Med. Sci. Innov. Hub., Riken*)

1Pos024* MARTINI 力場を用いた粗視化シミュレーションによる分子シャペロニン GroEL の ATP に誘起される構造変化の解析
Analysis of the ATP-induced conformational change of the molecular chaperonin GroEL by coarse-grained simulations using the MARTINI
Yuya Yamaura, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech*)

1Pos025* A Theoretically Study of ATP Effect on Solubility of Intrinsically Disordered Protein under Crowded Environment
Hayato Aida[1], Ryuhei Harada[2], Yasuteru Shigeta[2] ([1]*Coll. Bio. Sci., Univ. Tsukuba*, [2]*CCS, Univ. Tsukuba*)

1Pos026* 脂質膜表面におけるシトクロム P450 還元酵素の誘電アロステリー
Dielectric allostery in cytochrome P450 reductase on the surface of lipid membrane
Mikuru Iijima, Jun Ohnuki, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

1Pos027* TypeIII 中間径フィラメントの細胞表面上への出現機構の解明
Elucidation of recruitment mechanism of type III intermediate filament proteins to cell surface
Beomju Hwang[1], Inu Song[1], Hirohiko Ise[2] ([1]*Grad. Sch. Eng., Kyushu univ.*, [2]*IMCE, Kyushu Univ.*)

1Pos028* 量子化学計算による EcoRV のシシルリン酸基ツイスト後のリン酸エステル加水分解反応の研究
DNA Hydrolysis by EcoRV Subsequent to Scissile-Phosphate Twist, Studied by QM/MM Metadynamics Simulation
Itaru Onishi[1], Norio Yoshida[2], Fumio Hirata[3,4], Masayuki Irisa[1] ([1]*Kyushu Inst. of Tech.*, [2]*Kyushu Univ.*, [3]*IMS*, [4]*Toyota Riken*)

1Pos029 Crystal structure of branched-chain polyamine synthase
Eiichi Mizohata[1,2], Masataka Toyoda[1], Ryota Hidese[3], Shinsuke Fujiwara[3] ([1]*Grad. Sch. Eng., Osaka Univ.*, [2]*JST-PRESTO*, [3]*Grad. Sch. Sci. Tech., Kwansei Gakuin Univ.*)

1Pos030 Analysis of the complex molecular system composed of GGA, MPR and Ub by using titration SAXS measurement

Yugo Hayashi[1], Natsumi Endo[1], Youichi Yamazaki[1], Sachiko F. Toma[1], Hironari Kamikubo[1,2] ([1]*Div. Mat. Sci., NAIST*, [2]*IMSS KEK*)

1Pos031 Structural analysis of firefly luciferase with MM and QM/MM molecular simulations to clarify the origin of emission color-change factors

Kota Nosaka, Naohisa Wada (*Grad. Sch. Life Sci., Univ.Toyo*)

1Pos032 シアノバクテリア概日時計における KaiC 六量体の構造多様性
Structural Diversity of KaiC Hexamer in Cyanobacterial Circadian Clock

Yoshihiko Furuike[1,2], Atsushi Mukaiyama[1,2], Eiki Yamashita[3], Takao Kondo[4], Shuji Akiyama[1,2] ([1]*Research Center of Integrative Molecular Systems (CIMoS), Institute for Molecular Science (IMS)*, [2]*Department of Functional Molecular Science, SOKENDAI (The Graduate University for Advanced Studies)*, [3]*Institute for Protein Research, Osaka University*, [4]*Graduate School of Science, Nagoya University*)

1Pos033 NMR analysis of metal ion-induced conformational changes of α-helical peptides

Ikuko Iizumi[1], Yohei Miyanoiri[2], Toshiki Tanaka[3], Masayuki Oda[1] ([1]*Grad. Sch. Life Environ. Sci., Kyoto Pref. Univ.*, [2]*Inst. Protein Res., Osaka Univ.*, [3]*Nagoya Inst. Technol.*)

1Pos034 シクロスポリン A の CHARMM 力場の開発と膜—水系の分子動力学シミュレーションへの応用
Development of the CHARMM force field for Cyclosporine A and application to molecular dynamics simulations using a membrane-water system

Tsutomu Yamane[1], Ryo Takahashi[1], Toru Ekimoto[1], Mitsunori Ikeguchi[1,2] ([1]*Grad. Sch. Med. Life Sci, Yokohama City Univ.*, [2]*RIKEN Med. Sci. Innov. Hub*)

1Pos035 ヒト成人ヘモグロビンの酸素親和性制御に関連した GHz, THz 領域振動の研究
Study on Giga- and Terahertz-frequency Motions Involved with Oxygen Affinity of Human Adult Hemoglobin

Shigenori Nagatomo[1], Kohji Yamamoto[2], Masako Nagai[3], Teizo Kitagawa[4] ([1]*Dept. Chem., Univ. Tsukuba*, [2]*Res. Center Develop. Far-IR Region, Univ. Fukui*, [3]*Res. Center Micro-Nano Tech., Hosei Univ.*, [4]*Grad. Sch.Life Sci., Univ. Hyogo*)

1Pos036 Structural basis for the intramolecular signal transduction of oxygen sensor protein FixL from Bradyrhizobium japonicum

Misaki Kamaya[1], Hiroyasu Koteishi[1], Takehiko Tosha[1,2], Seiki Baba[3], Hiroshi Sugimoto[1,2], Yoshitsugu Shiro[1], Hitomi Sawai[1,2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*RIKEN SPring-8*, [3]*SPring-8 / JASRI*)

1Pos037 心筋ナトリウムチャネル Nav1.5 と薬剤間の結合自由エネルギー計算
Calculation of the binding free energies between the Nav1.5 sodium channel and drug molecules

Tatsuki Negami[1], Tohru Terada[2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

1Pos038 DNA 修復にかかわる Hef の天然変性領域の構造と機能
The structure and function of intrinsically disordered region of Hef that is associated with a DNA repair

Takashi Oda[1], Ayako Sekino[1], Ayaka Murakami[1], Rika Oi[1], Maki Yoneyama[1], Noriyuki Kodera[2], Toshio Ando[2], Tsuyoshi Konuma[3], Kenji Sugase[3], Tomotaka Oroguchi[4], Sonoko Ishino[5], Yoshizumi Ishino[5], Mamoru Sato[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City Unv.*, [2]*WPI NanoLSI, Kanazawa Univ.*, [3]*Grad.Sch. of Eng., Kyoto Univ.*, [4]*Faclt. Sci. Tech., Keio Univ.*, [5]*Grad. Sch. of Bioresource & Bioenviron. Sci., Kyushu Univ.*)

1Pos039 T2-like ファージ宿主認識蛋白質と宿主 OmpC の相互作用解析
Structural and functional analysis of phage receptor binding protein and OmpC

Shuji Kanamaru (*Dep. of Life Sci. & Tech., Tokyo Inst. of Tech.*)

1Pos040 ヤナギマツタケ（Agrocybe cylindracea）の Pri3 遺伝子のクローニングと特性解析
Cloning and characterization of the Pri3 gene of the edible mushroom, Agrocybe cylindracea

Chika Abematsu[1], Yamato Kuratani[1], Masashi Shin[1], Makoto Iwata[2], Toshihiko Matsumoto[1], Shoji Ando[1] ([1]*Fac. Biotech. Life Sci., Sojo Univ.*, [2]*IMB*)

1Pos041 ヒトヘアケラチン K85 の遺伝子導入細胞における機能特性と外胚葉形成不全症の原因となる変異の影響
Functional characteristics of human hair keratin K85 in transfected cells and the effects of mutations causative of ectodermal dysplasia
Masaki Yamamoto[1], Yasuko Sakamoto[1], Yuko Honda[2], Kenzo Koike[3], Hideaki Nakamura[4], Toshihiko Matsumoto[1], Shoji Ando[1] ([1]*Fac. Biotech. Life Sci., Sojo Univ.*, [2]*Fac. Med., Saga Univ.*, [3]*Kao corp.*, [4]*Fac. Phar., Sojo Univ.*)

1Pos042 巨大ヘモグロビンのアロステリック中間体の時分割構造解析
Time-resolved structure analysis of allosteric intermediate of the giant hemoglobin
Nobutaka Numoto[1], Yoshihiro Fukumori[2], Kunio Miki[3], Nobutoshi Ito[1] ([1]*Med. Res. Inst., Tokyo Med. Dent. Univ. (TMDU)*, [2]*College Sci. Eng., Kanazawa Univ.*, [3]*Grad. Sch. Sci., Kyoto Univ.*)

蛋白質：物性・構造／Protein: Property & Structure

1Pos043* 分子動力学法による RvSAHS1 の構造安定性
Structural stability of RvSAHS1 by MD simulations
Kazuhisa Miyazawa[1,2,3], Satoru Itoh[1,2,3], Hisashi Okumura[1,2,3] ([1]*SOKENDAI*, [2]*IMS*, [3]*ExCELLS*)

1Pos044* Rheo-NMR 法によるスーパーオキシドジスムターゼ 1 の動的なアミロイド形成機構の解析
Dynamic Analysis of Amyloid Formation of Superoxide Dismutase 1 Using Rheo-NMR Spectroscopy
Naoto Iwakawa[1], Daichi Morimoto[1], Erik Walinda[2], Masahiro Shirakawa[1], Kenji Sugase[1] ([1]*Grad. Sch. Eng., Kyoto Univ.*, [2]*Grad. Sch. Med., Kyoto Univ.*)

1Pos045* 統計力学モデルの拡張によるタンパク質のフォールディング反応機構の予測
Prediction of protein folding mechanisms by an extended statistical mechanical model
Koji Ooka[1], Munehito Arai[1,2] ([1]*Dept. Phys., Univ. Tokyo*, [2]*Dept. Life Sci., Univ. Tokyo*)

1Pos046 低波数基準振動で特徴づけられた多量体蛋白質の動的性質
Dynamic properties of oligomeric proteins characterized by low-frequency normal modes
Hiroshi Wako[2], **Shigeru Endo**[1] ([1]*Dept. of Phys., Sch. of Sci., Kitasato Univ.*, [2]*Sch. of Social Sci., Waseda Univ.*)

1Pos047 粗視化 Go モデルを用いた GA・GB ドメイン関連タンパク質のフォールディング機構の相違・共通性の予測
Prediction of differences and commonality in folding mechanisms of GA / GB domain related proteins using coarse-grained Go model
Shoya Hamaue (*Dept. of Bioinfo., Col. of Life sci., Ritsumeikan Univ.*)

1Pos048 Mechanism of the spontaneous elongation of the fibroin nanofiber involved in spider silk
Takuya Sawai[1], Kiichi Hayashi[1], Yugo Hayashi[1], Takehiro Sato[2], Yoichi Yamazaki[1], Sachiko Toma-Fukai[1], Hironari Kamikubo[1,3] ([1]*Div. Mat. Sci., NAIST*, [2]*Spiber Inc.*, [3]*IMSS KEK*)

1Pos049 天然様階層構造を有する人工クモ糸材料の再構成
Reconstitution of artificial materials of spider silk accompanied by native-like hierarchical structure
Satoru Onishi[1], Yuki Nakatani[1], Yugo Hayashi[1], Takehiro Sato[2], Yoichi Yamazaki[1], Sachiko Toma-Fukai[1], Hironari Kamikubo[1,3] ([1]*Div. Mat. Sci., NAIST*, [2]*Spiber Inc.*, [3]*IMSS KEK*)

蛋白質：機能／Protein: Function

1Pos050* ヒト SOD1 と Zn 欠損 SOD との比較による亜鉛と静電ポテンシャルループの役割に関する研究
Investigation on role of zinc atom and electrostatic loop by comparing human SOD1 with Zn-deficient SOD
Natsumi Koyama[1], Masami Lintuluoto[1], Juha Lintuluoto[2] ([1]*Grad. Sch. Life and Environ. sci., Kyoto pref. univ.*, [2]*Grad. sch. eng., Kyoto univ.*)

1Pos051 点変異導入による CD44 のヒアルロン酸結合のアロステリック制御
Allosteric regulation of hyaluronan binding on CD44 by point mutation
Masami Lintuluoto[1], Youta Horioka[1], Katsuhisa Matsumoto[1], Saki Hongo[1], Juha Lintuluoto[2] ([1]*Grad. Sch. Life and Environ. Sci., Kyoto pref. univ.*, [2]*Grad. sch. eng., Kyoto Univ.*)

1Pos052 アミロイド β42 を用いた天然変性タンパク質の分子シールド効果の評価
The evaluation of molecular shield effect of intrinsically disordered proteins using amyloid beta 1-42
Koki Ikeda, Yoshiki Shigemitsu, Natsuko Tenno, Takeshi Tenno, Hidekazu Hiroaki (*Grad. Sch. Pharm Sci., Univ. Nagoya*)

1Pos053 X 線小角散乱を用いた大腸菌フェリチンの鉄コアの形成に関する研究
The iron core formation of E. coli ferritin studied by small angle X-ray scattering
Takumi Kuwata, Daisuke Sato, Kazuo Fujiwara, Masamichi Ikeguchi (*Department of Bioinformatics, Soka University*)

1Pos054 柔らかいドライブシャフトを持つ F_1-ATPase のトルク伝達/発生
Torque transmission/generation of F_1-ATPase with a soft driveshaft
Shou Furuike[1], Naoki Soga[2], Yasushi Maki[1], Hideji Yoshida[1] ([1]*Phys., Osaka Med. Coll.*, [2]*Grad. Sch. Eng., Univ. Tokyo*)

蛋白質：計測・解析／Protein: Measurement & Analysis

1Pos055* (1SDP-3) Biophysical analysis of alpha-synuclein oligomers by microchip electrophoresis
William E. Arter[1,2], Catherine K. Xu[1], Georg Krainer[1], Christopher M. Dobson[1], Tuomas P. J. Knowles[1,2] ([1]*Centre for Misfolding Disease, Department of Chemistry, University of Cambridge*, [2]*Cavendish Laboratory, Department of Physics, University of Cambridge*)

1Pos056* タンパク質内部の構造変化をプローブするためのアスパラギン酸マッピングと赤外分光解析
Mapping of aspartic acids to probe protein structural changes by FTIR spectroscopy
Masanori Hashimoto, Kota Katayama, Manish Singh, Yuji Furutani, Hideki Kandori (*Grad. Sch. Eng., Nagoya Inst. Tech.*)

1Pos057* バイオ医薬品中の蛋白質凝集体の定量手法の確立
Establishment of quantification methods for protein aggregates in biopharmaceuticals
Saki Yoneda[1], Bertram Niederleitner[2], Michael Wiggenhorn[2], Hiroki Koga[1], Shinichiro Totoki[3], Elena Krayukhina[1], Wolfgang Friess[4], Susumu Uchiyama[1,5] ([1]*Dept. biotech. grad. sch. eng., Osaka. univ*, [2]*Coriolis Pharma*, [3]*Shimadzu Corporation*, [4]*LMU, Dept.Phaemacy*, [5]*ExCELLS*)

1Pos058 (1SDP-6) 新規に開発した高濃度タンパク質のためのネガティブ染色電子顕微鏡法
(1SDP-6) A newly developed negative stain EM method for protein complexes at high protein concentration
Hiroshi Imai[1], Takayuki Kato[2], Gerle Christoph[3], Etsuko Muto[4], Kaoru Mitsuoka[5], Genji Kurisu[3], Keiichi Namba[2], Takahide Kon[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*IPR, Osaka Univ.*, [4]*CBS, RIKEN*, [5]*Res. Ctr. UVHEM, Osaka Univ.*)

1Pos059 模倣細胞内分子混雑環境及び糖ガラス中の特定のタンパク質及び脂質膜の構造の中性子散乱による研究
Study of protein or membrane structure in mimicking intra-cell molecular-crowding environment and in sugar-glass using neutron scattering
Mitsuhiro Hirai[1], Satoshi Ajito[1], Shigeki Arai[2], Shinichi Takata[3], Hiroki Iwase[4] ([1]*Graduate School of Science and Technology, Gunma University*, [2]*National Institute for Quantum and Radiological Science and Technology*, [3]*J-PARC Center, Japan Atomic Energy Agency*, [4]*Comprehensive Research Organization for Science and Society*)

1Pos060 TRPV1 分子内部の回転動態の決定
Agonist- and antagonist-induced rotational motion of TRPV1 channel
Shoko Fujimura[1], Kazuhiro Mio[1], Masahiro Kuramochi[2], Sekiguchi Hiroshi[3], Muneyo Mio[1], Tai Kubo[1], Yuji Sasaki[1,2,3] ([1]*Operand OIL, National Institute of Advanced Industrial Science and Technology, Tokyo, Japan*, [2]*Graduate School of Frontier Sciences, The Univ. Tokyo, Chiba, Japan*, [3]*Japan Synchrotron Radiation Research Institute, Hyogo, Japan*)

1Pos061 レプリカ部分置換法の開発とタンパク質への応用
Development of replica sub-permutation method and its application to mini-protein
Masataka Yamauchi[1,2,3], Hisashi Okumura[1,2,3] ([1]*SOKENDAI*, [2]*ExCELLS*, [3]*IMS*)

1Pos062 凝縮系の不均一動力学(dynamical disorder)の分子論開拓: タンパク質の構造遷移・揺らぎ階層性
Theoretical investigations on microscopic heterogeneity and hierarchy in transitions and fluctuations of protein conformations
Yoshihiro Matsumura[1], Shinji Saito[1,2] ([1]*IMS*, [2]*SOKENDAI*)

1Pos063 Lytic polysaccharide monooxygenase の生化学および 1 分子解析
Biochemical and single-molecule analyses of lytic polysaccharide monooxygenase
Siti Mastura Zakaria[1,2], Akihiko Nakamura[1,3], Yasuko Okuni[1], Mayuko Yamamoto[1], Akasit Visootsat[1,3], Jun Ando[1,3], **Ryota Iino**[1,3] ([1]*IMS, NINS*, [2]*Univ. of Malaya*, [3]*SOKENDAI*)

蛋白質工学／Protein: Engineering

1Pos064 Relationship between loop geometry and register shift in parallel beta-sheet proteins
Ryuichiro Ueda, George Chikenji (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

1Pos065 2 アミノ酸同時変異戦略を用いた SBDD による超高親和性抗体の創製
Structure-based discovery of the antibodies with sub-picomolar affinity using the double amino acid mutation strategy
Shuntaro Chiba[1], Masateru Ohta[1], Aki Tanabe[2], Makoto Nakakido[2], Kouhei Tsumoto[2,3], Yasushi Okuno[1,4] ([1]*MIH, RIKEN*, [2]*Sch. Eng., Univ. Tokyo*, [3]*Inst. Med. Sci., Univ. Tokyo*, [4]*Grad. Sch. Med., Kyoto Univ.*)

1Pos066 転写コアクチベータ CBP の KIX ドメインと転写因子間の相互作用を阻害するペプチドの合理的設計
Rational design of peptides that inhibit interactions between the KIX domain of CBP and transcription factors
Nao Sato[1], Shunji Suetaka[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

1Pos067 分子動力学シミュレーションを用いた CDR-Grafting による合成単ドメイン抗体の構造的変化の解析
Structural effects of CDR grafting of synthetic single domain antibodies investigated by molecular dynamics simulations
Seisho Kinoshita[1], Chinatsu Mori[2], Makoto Nakakido[1,2], Daisuke Kuroda[1,2,3], Jose Caaveiro[4], Kouhei Tsumoto[1,2,3,5] ([1]*Dept. of Bioeng., Sch. of Eng., Univ. of Tokyo*, [2]*Dept. of Chem. Biotech., Sch. of Eng., Univ. of Tokyo*, [3]*Med. Dev. Dev. Reg. Res. Center, Sch. of Eng., Univ. of Tokyo*, [4]*Grad. Sch. of Pharm. Sci., Kyushu Univ.*, [5]*Inst. of Med. Sci., Univ. of Tokyo*)

1Pos068 アレルギー性喘息に関わるインターロイキン 33 の阻害剤開発に向けて
Toward the development of an inhibitor of interleukin-33 responsible for allergic asthma
Mio Sano[1], Yoshiki Oka[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

1Pos069 Domain-specific monitoring of higher-order structure of therapeutic IgG on the basis of molecular recognition of artificial proteins
Hideki Watanabe[1], Naoko Hayashida[2], Shinya Honda[1,2] ([1]*BMRI, AIST*, [2]*Grad. Sci. of Fro., Univ. of Tokyo*)

1Pos070 主鎖環状化が顆粒球コロニー刺激因子に与える会合体抑制効果
An effect of resistance to self-association of backbone circularization on granulocyte-colony stimulating factor
Risa Shibuya[1], Takamitsu Miyafusa[2], Shinya Honda[1,2] ([1]*Grad. Sch. of Fro., Univ. of Tokyo*, [2]*BMRI, AIST*)

ヘム蛋白質／Heme proteins

1Pos071 Function of N-terminal acetylation of fish hemoglobin α-subunit
Satoru Unzai, Antonio Tsunesige (*Hosei University, Department of Frontier Bioscience*)

1Pos072 Functional roles of conserved residues near the active site of nitric oxide reductase based on the structural analysis
Takehiko Tosha[1], Raika Yamagiwa[2], Takuya Kurahashi[2], Hiroshi Sugimoto[1], Yoshitsugu Shiro[2] ([1]*RIKEN SPring-8*, [2]*Univ. of Hyogo*)

1Pos073 2—状態アロステリーモデルの再検討ー機能的階層の有無
INSIGHT INTO THE TWO-STATE ALLOSTERIC MODEL - ON THE EXISTENCE OF HIERARCHIES
Antonio Tsuneshige, Satoru Unzai (*Hosei Univ. Frontier Bioscience*)

膜蛋白質／Membrane proteins

1Pos074* (1SCA-2) 脂質分子の混み合い効果による膜貫通タンパク質結晶化の検討
(1SCA-2) Crystallization of transmembrane protein driven by molecular crowding effect of lipids: Theoretical estimation by using a simple model
Keiju Suda[1], Ayumi Suematsu[2], Ryo Akiyama[1] ([1]*Kyushu University, Sci.*, [2]*Kyushu Sangyo University, Science and Engineering*)

1Pos075* アミロイド前駆体タンパク質と β 切断酵素の膜貫通部位の生体膜中での相互作用
Interaction between amyloid precursor protein and beta-secretase in the bio-membrane
Kaori Yanagino, Naoyuki Miyashita (*BOST KINDAI*)

1Pos076* 多次元固体 NMR による細胞膜中のヘリオロドプシンの構造解析
Multidimensional solid-state NMR study of heliorhodopsin in a lipid environment
Shibuki Suzuki[1], Toshio Nagashima[2], Rina Kaneko[1], Takashi Okitsu[3], Akimori Wada[3], Naohiro Kobayashi[2], Toshio Yamazaki[2], Rei Abe-Yoshizumi[4], Keiichi Inoue[5], Hideki Kandori[4], Izuru Kawamura[1] ([1]*Grad. Sch. Eng. Sci., Yokohama National Univ.*, [2]*RIKEN RSC*, [3]*Kobe Pharmaceutical Univ.*, [4]*Nagoya Inst. Tech.*, [5]*Univ. Tokyo*)

1Pos077 (1SHP-6) 理論計算による熱安定化ムスカリン M2 受容体の選択的アンタゴニスト AF-DX 384 結合型構造
(1SHP-6) Structural insights into the subtype-selective antagonist binding to the M2 muscarinic receptor
Ryoji Suno[1], Sangbae Lee[2], Shoji Maeda[3], Satoshi Yasuda[4], Keitaro Yamashita[9], Kunio Hirata[5,6], Takeshi Murata[7], Masahiro Kinoshita[8], Masaki Yamamoto[5], Brian Kobilka[3], Nagarajan Vaidehi[2], So Iwata[8], Takuya Kobayashi[1] ([1]*Kansai Medi. Univ.*, [2]*City Hope Med Ctr.*, [3]*Stanford Univ.*, [4]*Chiba Univ.*, [5]*RIKEN,SPring-8*, [6]*JST,PRESTO*, [7]*IAE, Kyoto Univ.*, [8]*Med,Kyoto Univ.*, [9]*Univ.Tokyo,Sci*)

1Pos078 (1SCA-4) cDNA ディスプレイとセルソーターの利用による新規リポソームポア形成ペプチドの創製
(1SCA-4) Novel pore-forming peptides assembling in liposome membranes selected by combining cDNA display method with cell sorter system
Naoto Nemoto[1], Toshiki Miyajima[1], Takeru Yoshinobu[1], Yusuke Sekiya[2], Ryuji Kawano[2] ([1]*Grad. Sci. Eng., Saitama Univ.*, [2]*Dept. Biotech. Life Sci., Tokyo Univ. Agr. Tech*)

1Pos079 特異的な残基置換が、水和水のダイナミクスにおよぼす影響を、解析するソフトを開発
Development of software to analyze the effects of specific residue substitution on hydration water dynamics
Ryoi Ashida, Nobuya Hasegawa, Takuya Azami, Kota Kasahara, Takuya Takahashi (*Graduate School of Life Science, Ritsumeikan University*)

1Pos080 A comparative study of membrane-bound structure of antimicrobial peptides L- and D-phenylseptin
Izuru Kawamura[1], Batsaikhan Mijiddorj[1], Hisako Sato[2], Yuta Matsuo[1], Akira Naito[1], Kazuyoshi Ueda[1] ([1]*Grad.Sch. Eng., Yokohama Natl. Univ.*, [2]*Grad. Sch. Sci. Eng.*)

核酸結合蛋白質／Nucleic acid binding proteins

1Pos081* Observation of Tumor Suppressor p53 Search Dynamics using Sub-millisecond Resolved Single-molecule Fluorescence Microscopy
Dwiky Rendra Graha Subekti[1,2], Agato Murata[1], Yuji Itoh[1], Satoshi Takahashi[1], Kiyoto Kamagata[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Sci., Tohoku Univ.*)

1Pos082 (1SBA-6) 大腸菌非六量体型 DNA ヘリカーゼ UvrD C 末端欠損変異体の 1 分子イメージング
(1SBA-6) Single-molecule imaging of a non-hexameric *Escherichia coli* helicase UvrD mutant lacking C-terminal residues
Hiroaki Yokota (*Biophotonics Lab., Grad. Sch. Creation New Photon. Indust.*)

1Pos083 スピンラベル ESR によるヘテロクロマチンタンパク質 HP1 の動的構造の研究：アイソフォーム特異性とリン酸化
Structural dynamics of heterochromatin protein HP1 studied by spin labeling ESR spectroscopy: Isoform specificity and phosphorylation
Toshiaki Arata[1,5], Shigeaki Nakazawa[4], Yuichi Mishima[5], Kazunobu Sato[4], Takeji Takui[4], Toru Kawakami[5], Hironobu Hojo[5], Toshimichi Fujiwara[5], Makoto Miyata[1], Isao Suetake[2,3,5] ([1]*Dept. Biol., Grad. Sch. Sci., Osaka City Univ.*, [2]*Koshien Univ.*, [3]*Twin Research Center, Osaka Univ.*, [4]*Dept. Chem., Grad. Sch. Sci., Osaka City Univ.*, [5]*IPR, Osaka Univ.*)

1Pos084 Protein localization in DNA cruciform junction studied by molecular simulation
Mami Saito, Shoji Takada (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

1Pos085 HIV Vif–ヒト E3 ユビキチンリガーゼ複合体によるヒト抗ウイルス因子 APOBEC3G の脱アミノ化阻害の分子メカニズム
Inhibition mechanism of HIV Vif-human E3 ubiquitin ligase complex against enzymatic activity of APOBEC3G
Keisuke Kamba[1], Li Wan[1,2], Satoru Unzai[3], Ryo Morishita[4], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Inst. of Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Energy Sci., Kyoto Univ.*, [3]*Front. Biosci., Hosei Univ.*, [4]*CellFree Sciences Co.,Ltd.*)

核酸／Nucleic acid

1Pos086* 薬剤によるリボスイッチ“SPINACH”の構造とイオンへの影響
Influence on the structure and dynamics of Riboswitch “SPINACH” and potassium ions by DFHBI
Lisa Matsukura[1], Nobutaka Onishi[1], Masaya Furue[1], Naoyuki Miyashita[1], Takuma Shiraki[1], Yasushige Yonezawa[2] ([1]*BOST, KINDAI*, [2]*IAT, KINDAI*)

1Pos087* シスプラチンは二本鎖 DNA を桁違いに硬くする：一分子揺らぎの定量的解析
Cisplatin causes DNA much stiffer: Quantitative evaluation viscoelasticity through the analysis of single molecule fluctuation
Toshifumi Kishimoto[1], Yuko Yoshikawa[1], Seiji Komeda[2], Kenichi Yoshikawa[1] ([1]*Grad.Sch.Life Med.Sci.,Univ.Doshisha*, [2]*Fac.Pharm.,Univ.Suzuka.Med.Sci*)

1Pos088 (1SEA-6) エピジェネティック修飾をもつクロマチンのモデルにおける不連続相転移
(1SEA-6) Discontinuous Phase Transition in a Chromatin Model with Epigenetic Modification
Kyosuke Adachi, Kyogo Kawaguchi (*RIKEN BDR*)

1Pos089 トリヌクレオソームから構築するポリヌクレオソーム構造の特徴
Characterization of Poly-nucleosome Structure Constructed from Tri-nucleosome
Hiroo Kenzaki[1], Shoji Takada[2] ([1]*Info. Sys. Div., ISC, RIKEN*, [2]*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ.*)

1Pos090 局所変形下における絡まりあった DNA 溶液の緩和過程
Relaxation process of entangled DNA solution after local deformation
Akinori Miyamoto, Yoshihiro Murayama (*Department of Applied Physics,Tokyo University of Agri. and Tech.*)

1Pos091 In-cell NMR 法を用いたヒト生細胞内核酸の構造およびダイナミクスの評価
Evaluation of the structure and dynamics of nucleic acids inside the living human cells by in-cell NMR spectroscopy
Yudai Yamaoki[1], Takashi Nagata[1,2], Ayaka Kiyoishi[2], Masayuki Miyake[2], Fumi Kano[3], Masayuki Murata[3,4], Masato Katahira[1,2] ([1]*Inst. Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Energy Sci., Kyoto Univ.*, [3]*Inst. Innovative Res., Tokyo Inst. Technol.*, [4]*Grad. Sch. Arts and Sci., Univ. Tokyo*)

1Pos092 ハンマーヘッドリボザイムの酵素反応に関する理論的研究
Theoretical study on an enzymatic reaction of the hammerhead ribozyme
Ayaka Matsuyama, Masahiko Taguchi, Shigehiko Hayashi (*Kyoto University*)

1Pos093* 2 価ポリアミンが引き起こす遺伝子発現の促進と抑制：アミノ基間の炭素鎖長の重要性
Promotion and inhibition of gene expression caused by divalent polyamines: Marked effect of the distance between amino groups
Hiroko Tanaka[1], Chwen-Yang Shew[2], Yuko Yoshikawa[1], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life. Med. Sci., Univ. Doshisha*, [2]*Col. Chem., City Univ. New York*)

1Pos094* Theoretical Studies on the Conformational Stability of RA-VII complex with 60S Ribosome
Arwansyah MS[1], Yoh Noguchi[2], Takeshi Miyakawa[2], Kazutomo Kawaguchi[1], Yukio Hitotsuyanagi[3], Satoshi Yokojima[3], Ryota Morikawa[2], Masako Takasu[2], Hidemi Nagao[1] ([1]*Division of Mathematical and Physical Sciences, Graduate School of Natural Science and Technology, Kanazawa University*, [2]*School of Life Science, Tokyo University of Pharmacy and Life Sciences*, [3]*School of Pharmacy, Tokyo University of Pharmacy and Life Sciences*)

1Pos095 (1SEA-3) オリゴペプチドのアミノ酸配列は DNA compaction と転写活性に著しい違いを引き起こす
(1SEA-3) Marked Difference in DNA Compaction and Transcription is Caused by Amino Acid Sequence of Oligopeptide
Tatsuo Akitaya[1], Hiroyuki Hiramatsu[2], Hideaki Yamaguchi[3], Koji Kubo[4], Shizuaki Murata[4], Toshio Kanbe[5], Norio Hazemoto[6], Kenichi Yoshikawa[7], Anatoly Zinchenko[4] ([1]*Asahikawa Med. Univ.*, [2]*Fac. Pharm., Meijo Univ.*, [3]*Fac. Agr. Sci., Meijo Univ.*, [4]*Grad. Sch. Env. Std., Nagoya Univ.*, [5]*Grad. Sch. Med., Nagoya Univ.*, [6]*Grad. Sch. Pharm. Sci., Nggoya City Univ.*, [7]*Fac. Bio. Med. Sci., Doshisah Univ.*)

電子状態／Electronic state

1Pos096 Long-range Electron-Electron Interaction and Charge Transfer in Protein Complexes
David Gnandt, Thorsten Koslowski (*Institute of Physical Chemistry, University of Freiburg*)

1Pos097 —

1Pos098 拘束条件を用いた DFTB-MD シミュレーションの高速化とエネルギー保存の評価
Accelerate simulations and assessment of the energy conservation for the DFTB-MD simulations using the constraint method
Hiroaki Nishizawa, Yasuteru Shigeta (*CCS, Univ. of Tsukuba*)

1Pos099 光合成系 II の酸素発生中心の S1 状態での 12 の構造モデルの DLPNO-CCSD(T)法による計算
DLPNO-CCSD(T) calculations of twelve structural models for the S1 state of oxygen evolving complex of photosystem II
Koichi Miyagawa[1], Takashi Kawakami[2,5], Hiroshi Isobe[3], Mitsuo Shoji[4], Shusuke Yamanaka[2], Kazuhiko Nakatani[1], Mitsutaka Okumura[2], Takahito Nakajima[5], Kizashi Yamaguchi[1,5,6] ([1]*ISIR, Osaka Univ.*, [2]*Grad. Sch. of Sci., Osaka Univ.*, [3]*Grad. Sch. of Nat. Sci. and Tech., Okayama Univ.*, [4]*CCS, Univ. of Tsukuba*, [5]*R-CCS, RIKEN*, [6]*Inst. Nanosci. Design, Osaka Univ.*)

水・水和／電解質／Water & Hydration & Electrolyte

1Pos100* 生体溶液中のアニオンサイト間実効引力が発生する最大のサイトサイズ
Maximum size for attractive anionic-sites in a biological solution
Michika Takeda[1], Ayumi Suematsu[2], Ryo Akiyama[3] ([1]*Graduate of Science, Kyushu University*, [2]*Kyushu Sanyo University*, [3]*Institute of Science, Kyushu University*)

1Pos101 MD シミュレーションで、モデルタンパク質の水和水のダイナミクスを明らかにする
MD simulations reveal hydration dynamics around model proteins
Takuya Takahashi[1], Takuya Azami[2], Nobuya Hasegawa[1], Ryoi Ashida[1,2], Kota Kasahara[1] ([1]*Coll. Life Sci., Ritsumeikan Univ.*, [2]*Grad. Sch. Life Sci., Ritsumeikan Univ*)

1Pos102 All-atom molecular dynamics simulation of the reduced protein-protein interaction with metabolites in the cytoplasm
Isseki Yu[1,2], Michael Feig[3], Yuji Sugita[2] ([1]*Maebashi Institute of Technology*, [2]*RIKEN Theoretical Molecular Science Lab.*, [3]*Michigan State University*)

1Pos103 プロテインーリガンド結合における水溶媒の役割
Role of water solvent in protein-ligand binding
Yutaka Maruyama[1], Ayori Mitsutake[2], Takefumi Yamashita[3] ([1]*RIKEN R-CCS*, [2]*Dep. Phys., Meiji Univ.*, [3]*LSBM, Univ. Tokyo*)

発生・分化／Development & Differentiation

1Pos104 Mechanical symmetry breaking in *C. elegans* dorsal-ventral axis establishment
Masatoshi Nishikawa (*Dept. Frontier Bioscience, Hosei Univ.*)

1Pos105 マウスノード不動繊毛はメカノセンサーか?: 光ピンセットによる機械刺激後の Ca^{2+}応答
Are the immotile nodal cilia in mouse embryo mechanosensors?: Ca^{2+} signaling response after mechanical stimulation by optical tweezers
Takanobu A Katoh, Katsutoshi Mizuno, Hiroshi Hamada (*BDR, Riken*)

1Pos106 ライブセルイメージングが切り開くシアノバクテリアのヘテロシスト分化空間パターン維持機構
Live cell imaging sheds light on the maintenance mechanism of pattern of cyanobacterial cell differentiation
Shun-ichi Fukushima[1], Takeharu Nagai[1], Shigeki Ehira[2] ([1]*The Institute of Scientific and Industrial Research, Osaka University*, [2]*Department of Biological Sciences, Graduate School of Science, Tokyo Metropolitan University*)

1Pos107 細胞性粘菌における既知のシグナルを用いない細胞分化状態の検出
Detection of cell differentiation states without known signals in *Dictyostelium*
Yusuke V. Morimoto[1,2], Takuro Kawada[1], Masahiro Ueda[2,3] ([1]*Fac. Comp. Sci. and Sys. Eng., Kyushu Inst. Tech.*, [2]*BDR, RIKEN*, [3]*Grad. Sch. Frontier Biosci., Osaka Univ.*)

筋肉／Muscle

1Pos108 (1SGA-2) 心筋細胞に備わる収縮リズム恒常性の分子機構の解明
(1SGA-2) Elucidation of molecular mechanism of contraction rhythm homeostasis in cardiac myocytes
Seine Shintani[1], Takumi Washio[2] ([1]*Department of Biomedical Sciences, College of Life and Health Sciences, Chubu University*, [2]*Graduate School of Frontier Sciences, the University of Tokyo*)

1Pos109 トロポニン T の E244D 変異による細いフィラメント構造変化の X 線小角散乱解析
Structural changes of cardiac thin filaments caused by E244D mutation of troponin T observed by small-angle X-ray scattering
Tatsuhito Matsuo, Satoru Fujiwara (*Inst. Quant. Life Sci., QST*)

1Pos110 分子動力学計算によるミオシンの第二リン酸結合部位の検証
Validation of second phosphate binding site in myosin studied by molecular dynamics simulation
Kouei Uchida, Jun Ohnuki, Takato Sato, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

1Pos111 ミオシンのアクチンに対する結合親和性の低温における弱化を説明するための統計熱力学
Statistical thermodynamics on the weakening of binding affinity of myosin for actin at low temperatures
Tomohiko Hayashi, Masahiro Kinoshita (*Inst. Adv. Energy, Kyoto Univ.*)

1Pos112 1 分子・多分子実験から迫る、心機能に適した心筋ミオシンの性質
Reverse stroke of cardiac myosin revealed by single molecule microscopy is essential for heart function
Yongtae Hwang[1], Takumi Washio[2], Hideo Higuchi[1], Motoshi Kaya[1] ([1]*Department of Physics, The University of Tokyo*, [2]*Department of Human and Engineered Environmental Studies, The University of Tokyo*)

分子モーター／Molecular motor

1Pos113* 負荷に依存した細菌べん毛モーター回転速度の調節機構
Load-dependent speed regulation of the bacterial flagellar motor
Tsubasa Ishida[1], Myu Yoshida[2], Tohru Minamino[3], Yoshiyuki Sowa[1,2,4] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [4]*Res. Cent. Micro-Nano Tech., Hosei Univ.*)

1Pos114* F_1-ATPase の阻害状態解析
Analysis of the inhibited form of F_1-ATPase
Sougo Mori, Hiroshi Ueno, Hiroyuki Noji (*Noji laboratory, Department of Applied Chemistry, Graduate School of Engineering, University of Tokyo*)

1Pos115* Na^+の存在に依存するべん毛モーター固定子 PomAB における複合体の解離
Destabilization of the complex formation allows high Na^+ conduction in the PomAB flagellar stator complex
Tatsuro Nishikino, Hiroto Iwatsuki, Taira Mino, Seiji Kojima, Michio Homma (*Div. Biol. Sci. Grad. Sch. Sci., Nagoya Univ.*)

1Pos116* Biomolecular motor driven cargo transportation by microtubules as a mechanosensor
Syeda Rubaiya Nasrin[1], Arif Md. Rashedul Kabir[2], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Grad. Sch. Chem. Sci. and Engg., Hokkaido Univ.*, [2]*Fac. Sci., Hokkaido Univ.*)

1Pos117* ハイブリッド F_1-ATPase の 1 分子回転観察
Rotation observation of hybrid F_1-ATPases between bacterial and mammalian ones
Ryo Watanabe[1], Hiroshi Ueno[1], Toshiharu Suzuki[2], Ryohei Kobayashi[1], Hiroyuki Noji[1] ([1]*Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*CLS., Tokyo tech.*)

1Pos118* 生体分子群ロボットによる光制御時空間貨物輸送
Photo-regulated spatiotemporal cargo transportation by biomolecular swarm robot
Mousumi Akter[1], Jakia Jannat Keya[2], Arif Md. Rashedul Kabir[2], Hiroyuki Asanuma[3], Kuzuya Akinori[4], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Graduate School of Chemical Science and Engineering, Hokkaido University, Sapporo 060-0810, Japan*, [2]*Faculty of Science, Hokkaido University, Sapporo 060-0810, Japan*, [3]*Department of Biomolecular Engineering, Nagoya University, Nagoya, Japan*, [4]*Department of Chemistry and Materials Engineering, Kansai University, Osaka 564-8680, Japan*)

1Pos119* べん毛モーターの分子軸受 LP リングのクライオ電子顕微鏡による構造解析
CryoEM structural analysis of the bacterial flagellar LP ring ~ a molecular bushing of the bacterial motor with almost no friction ~
Tomoko Yamaguchi[1,2], Fumiaki Makino[1,3], Tomoko Miyata[1], Takayuki Kato[1], Keiichi Namba[1,2,4] ([1]*Grad. Sch. FBS, Univ. Osaka*, [2]*BDR, Riken*, [3]*JEOL Ltd.*, [4]*Spring-8, Riken*)

1Pos120 べん毛フックの自然な構造
Structure of the native supercoiled flagellar hook
Takayuki Kato[1], Tomoko Miyata[1], Fumiaki Makino[1,2], Peter Horvath[1], Keiichi Namba[1,3] ([1]*Front. Bio., Osaka univ.*, [2]*JEOL*, [3]*BDR, Riken*)

1Pos121 細菌べん毛基部体の極低温電子顕微鏡による構造解析
Structure analysis of the bacterial flagellar basal body by electron microscopy
Tomoko Miyata[1], Takayuki Kato[1], Fumiaki Makino[1,2], Yumiko Saijo[3], Keiichi Namba[1,4] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*JEOL Ltd*, [3]*Grad. Sch. Med., Kobe Univ.*, [4]*BDR&SPring8, RIKEN*)

1Pos122 Structure of motor evolved by combination of F-ATPase and phosphoglycerate kinase for *Mycoplasma mobile* gliding
Takuma Toyonaga[1], Takayuki Kato[2], Akihiro Kawamoto[3], Tasuku Hamaguchi[4], Keiichi Namba[2,4,5], Makoto Miyata[1,6] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*, [3]*IPR, Osaka Univ.*, [4]*SPring-8, RIKEN*, [5]*BDR, RIKEN*, [6]*OCARINA, Osaka City Univ.*)

1Pos123 コンデンシン分子モーターの DNA カーテン測定と構造モデリング
DNA curtain assay and structural modeling of condensin molecular motor
Hiroki Koide, Shoji Takada, Tsuyoshi Terakawa (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

1Pos124 nanodisc-Vo の再構成条件検討と単粒子解析
The Investigation of reconstruction and Single Particle Analysis of nanodisc-Vo
Aya Furuta[1], Jun-ichi Kishikawa[1], Takayuki Kato[2], Atsuko Nakanishi[1], Kaoru Mitsuoka[3], Ken Yokoyama[1] ([1]*Div. Life Sci., Kyoto Sangyo Univ.*, [2]*Grad. Sch. Frontier Biosci.,Osaka Univ.*, [3]*Res. Ctr. UHVEM., Osaka Univ.*)

1Pos125 クシクラゲの櫛板はシンクロトロン放射光による繊毛軸糸の構造・機能解析に最適な試料である
Synchrotron radiation X-ray diffractionv reveals the highly ordered structure of axonemes in the comb plate of ctenophores
Hiroyuki Iwamoto[1], Kei Jokura[2], Kazuo Inaba[2] ([1]*SPring-8, JASRI*, [2]*Shimoda Marine Res. Ctr., Univ. Tsukuba*)

1Pos126 Unidirectional-rotation and helical-motion of a cargo on a microtubule indicate torque generation and biased sideward binding of kinesin
Mitsuhiro Sugawa, Yohei Maruyama, Masahiko Yamagishi, Junichiro Yajima (*Grad. Sch. Arts and Sciences, Univ. Tokyo*)

1Pos127 ウシミトコンドリア由来 F_1-ATPase の 1 分子回転解析
Single-molecule analysis of bovine mitochondrial F_1-ATPase
Ryohei Kobayashi, Hiroshi Ueno, Hiroyuki Noji (*Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*)

1Pos128 二つの頭部のつながり方がミオシン VI の歩行運動に与える影響
Effects of interhead connection on the stepping motion of myosin VI
Tomoki P. Terada[1], Qing-Miao Nie[2], Masaki Sasai[1] ([1]*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*, [2]*Dept. Appl. Phys., Zhejiang Univ. Tech.*)

1Pos129 Allosteric Regulation of V_1-ATPase by Designing Non-catalytic Interface
Takahiro Kosugi[1,2,3], Tatsuya Iida[2], Mikio Tanabe[4], Ryota Iino[1,2], Nobuyasu Koga[1,2,3] ([1]*Institute for Molecular Science*, [2]*SOKENDAI*, [3]*ExCELLS*, [4]*KEK*)

1Pos130 キネシン-1 人工多量体のプロセシビティと一方向運動性の評価
Evaluation of processivity and unidirectionality of artificial kinesin-1 oligomers
Kimitoshi Takeda, Akihiko Nakamura, Jun Ando, Ryota Iino (*Institute for Molecular Science*)

1Pos131 DNA ナノフィラメント上を移動する改変型ダイニンを用いた人工輸送システムの創生
Creating artificial transport systems by using engineered dynein that moves along DNA nanofilament
Ryota Ibusuki[1], Tatsuya Morishita[1], Akane Furuta[2], Kazuhiro Oiwa[1,2], Hiroaki Kojima[2], Ken'ya Furuta[2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICR. Res. Ins., NICT. Kobe*)

1Pos132 DNA 上を動く改変ダイニンを用いて結合性と運動速度の関係を系統的に調べる
Systematic studies on the relation between binding kinetics and speed of movement using engineered DNA-based dynein motor
Tatsuya Morishita[1], Ryota Ibusuki[1], Akane Furuta[2], Kazuhiro Oiwa[1,2], Hiroaki Kojima[2], Ken'ya Furuta[2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst NICT*)

1Pos133 Fo 回転モーターにおける固定子ー回転子間相互作用の自由エネルギー地形
Free energy landscape for stator-rotor interaction in Fo rotary motor
Dan Parkin, Daiki Yamakoshi, Genya Nakagawa, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

1Pos134 ミオシン頭部の自由エネルギーランドスケープのスイッチングを考慮した筋収縮の三状態モデル
Three-state model of muscle contraction with switched free energy landscapes for myosin heads
Kaima Matsuda, Masaki Sasai, Tomoki P. Terada (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

1Pos135 DNA オリガミを用いたミオシンフィラメントでのミオシン II モーターの高速原子間力顕微鏡による同時観察
Simultaneous observation of individual myosin II motors in DNA origami-based thick filaments by high-speed AFM
Masashi Ohmachi[1], Hiroaki Takagi[2], Keigo Ikezaki[3], Toshio Yanagida[1,4], Mitsuhiro Iwaki[1,4] ([1]*BDR, RIKEN*, [2]*Nara Med. Univ.*, [3]*Univ. Tokyo*, [4]*Grad. Sch. Front. Biosci., Osaka Univ.*)

1Pos136 アクトミオシン運動に対するリゾチームの阻害作用
Inhibitory effect of lysozyme on the motility of actomyosin
Masaki Okami, Kuniyuki Hatori (*Grad.Sch.Sci.Eng., Yamagata Univ.*)

1Pos137 ダイナクチンサイドアームのコンフォメーション変化
Multiple conformational changes of dynactin sidearm revealed by single molecule observation
Kei Saito[1], Takuya Kobayashi[2], Takashi Murayama[2], Mitsuhiro Sugawa[1], Christian Ganser[3], Takayuki Uchihashi[3,4], Junichiro Yajima[1], Yoko Y. Toyoshima[1] ([1]*Grad. Sch. Arts Sci., Univ. Tokyo*, [2]*Dept. of Pharmacology, Juntendo Univ. Sch. of Med.*, [3]*NINS, ExCELLS*, [4]*Dept. of Phys., Nagoya Univ.*)

1Pos138 The contribution of microtubule-binding ability of dynactin in dynein behaviors on microtubules
Takuya Kobayashi[1], Kei Saito[2], Takuya Miyashita[2], Yoko Y Toyoshima[2], Takashi Murayama[1] ([1]*Dept. Pharmacology, Juntendo Univ.*, [2]*Grad. sch. Arts and Sci. Univ. Tokyo*)

細胞生物的課題／Cell biology

1Pos139* (3SDA-6) 集団細胞遊走における機械的なシグナルを介した ERK 活性伝播
(3SDA-6) ERK activation waves mediated by intercellular mechanical signals during collective cell migration
Naoya Hino[1], Xavier Trepat[2], Michiyuki Matsuda[1,3], Tsuyoshi Hirashima[3] ([1]*Grad. Sch. of Biostudies, Kyoto Univ.*, [2]*IBEC, Spain*, [3]*Grad. Sch. of Med., Kyoto Univ.*)

1Pos140* スピロプラズマの螺旋交換遊泳を駆動する内部リボン構造
Internal ribbon structure driving helicity-switching swimming of *Spiroplasma*
Yuya Sasajima[1], Takayuki Kato[2], Tomoko Miyata[2], Keiishi Namba[2,3], Makoto Miyata[1,4] ([1]*Grad. Sch. Sci., Osaka City Univ., Japan*, [2]*Grad. Sch. Front. Biosci., Osaka Univ., Japan*, [3]*BDR & SPring-8 Center, Riken, Japan*, [4]*OCARINA, Osaka City Univ., Japan*)

1Pos141* 液-液相分離における荷電性残基の影響に関する分子動力学シミュレーション
Molecular dynamics simulations to dissect effects of charged residues on liquid-liquid phase separation
Hiroki Terazawa[1], Kota Kasahara[2], Takuya Takahashi[2] ([1]*Grad. Sch. Life Sci., Ritsumeikan Univ.*, [2]*Coll. Life Sci., Ritsumeikan Univ.*)

1Pos142* *Spiroplasma eriocheiris* の遊泳運動にかかわる細菌のアクチン MreB の重合
Polymerization of bacterial actin MreB involved in swimming motility of *Spiroplasma eriocheiris*
Daichi Takahashi[1], Makoto Miyata[1,2] ([1]*Grad. Sch. Sci, Osaka City Univ.*, [2]*OCARINA, Osaka City Univ.*)

1Pos143* クラミドモナス細胞の鞭毛打頻度を用いた細胞内 ATP 濃度の推定
Estimation of intracellular ATP concentration from the flagellar beat frequency in *Chlamydomonas*
Wakako Takano[1,2], Toru Hisabori[1,2], Ken-ichi Wakabayashi[1,2] ([1]*CLS, Tokyo Tech*, [2]*LST, Tokyo Tech*)

1Pos144* 細菌バイオフィムの高次秩序構造
High structural order in bacteria biofilms
Kohei Takahashi[1], Kana Morinaga[1,2], Masanonri Toyofuku[3,4], Utada Andrew[3,4] ([1]*Graduate school of Life and Environment Science, University of Tsukuba*, [2]*Bioproduction Research Institute, Advanced Industrial Science and Technology*, [3]*Faculty Life Environment Science, University of Tsukuba*, [4]*Microbiology Research Center for Sustainability (MiCS), University of Tsukuba*)

1Pos145* 合成細菌における *Spiroplasma eriocheiris* 遊泳運動の再現
Reconstitution of *Spiroplasma eriocheiris* swimming motility in a synthetic bacterium
Hana Kiyama[1], Shigeyuki Kakizawa[2], Makoto Miyata[1,3] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*AIST,Bioprocess*, [3]*OCARINA, Osaka City Univ.*)

1Pos146 (1SHA-3) Direct observation of cell mechanics under high hydrostatic pressure
Masatoshi Morimatsu, Keiji Naruse (*Grad. Sch. of Med., Dent. and Pharma. Sci., Okayama Univ.*)

1Pos147 (1SHA-2) RhoA activation induces cell cycle exit and differentiation of skin cancer cells
Oleg Dobrokhotov, Masahiro Sokabe, Hiroaki Hirata (*Nagoya Univ., Grad. Sch. Med.*)

1Pos148 ゼブラフィッシュ胚上皮組織に内在する力が創傷治癒を制御する
Residual Stress-mediated wound healing in zebrafish epithelia
Sohei Yamada[1], Yasumasa Bessho[2], Yoichiroh Hosokawa[1], Takaaki Matsui[2] ([1]*Division of Materials Science, Nara Institute of Science and Technology*, [2]*Division of Biological Science, Nara Institute of Science and Technology*)

1Pos149 TIRF 観察によるアクチン重合・脱重合における蛍光標識の影響
Effects of Dye Labels on Actin Assembly and Disassembly
Ikuko Fujiwara[1], Shuichi Takeda[2], Toshiro Oda[5], Thomas Pollard[3], Naomi Courtemanche[4], Akihiro Narita[2], Yuichiro Maeda[2] ([1]*Grad.Sch.Sci., Osaka City Univ.*, [2]*Structural Biol. Res. Cent. Gad. Sch.Sci., Nagoya Univ.*, [3]*MCDB, Yale Univ., USA*, [4]*Univ. Minnesota, USA*, [5]*Tokaigakuin Univ.*)

1Pos150 変異リアノジン受容体の分子動力学シミュレーション
Molecular dynamics simulation of mutant ryanodine receptors
Toshiko Yamazawa[1], Haruo Ogawa[2], Maki Yamaguchi[1], Takashi Murayama[3], Hideto Oyamada[4], Nagomi Kurebayashi[3], Junji Suzuki[5], Kazunori Kanemaru[6], Katsuji Oguchi[4], Takashi Sakurai[3], Masamitsu Iino[6] ([1]*Dept Mol. Physiol., Jikei Univ. Sch. Med.*, [2]*Institute Quantitative Biosci., The Univ.Tokyo*, [3]*Dept. Pharmacol., Juntendo Univ. Sch. Med.*, [4]*Dept. Pharmacol., Sch. Med., Showa Univ*, [5]*Dept. Physiol., Univ. California San Francisco*, [6]*Dept. Cell. Mol. Pharmacol., Nihon Univ. Sch. Med.*)

1Pos151 脱重合が引き起こす接着斑周囲におけるアクチン細胞骨格の向きの変化
The change of the direction of F-actin caused by the filament disassembly around focal adhesions
Kiyoshi Tohyama[1], Sawako Yamashiro[2], Naoki Watanabe[1,2] ([1]*Department of Pharmacology, Kyoto University Faculty of Medicine*, [2]*Laboratory of Single-Molecule Cell Biology, Kyoto University Graduate School of Biostudies*)

1Pos152 クラミドモナス鞭毛の根元に局在するマイナータイプダイニンの機能解析
Functional analysis of minor-type axonemal dyneins located to the basal region of *Chlamydomonas* flagella
Tomohiro Komatsu, Yusuke Kondoh, **Toshiki Yagi** (*Dept. Life Sci., Pref. Univ. Hiroshima*)

1Pos153 共焦点レーザー走査型顕微鏡で捉えた軸索タンパクのブラウン運動
Brownian motions of axonal proteins captured by a confocal laser scanning microscopy
Kazunari Mouri[1], Yasushi Okada[1,2] ([1]*RIKEN, BDR*, [2]*Univ. Tokyo, Grad. Sch. Sci., Dept. Phys.*)

1Pos154 生体内光架橋によるべん毛回転子タンパク質 FliG と固定子タンパク質 PomA 間相互作用の検出
Interaction between the flagellar rotor protein FliG and the stator protein PomA in cells detected by *in vivo* photo-crosslink
Seiji Kojima, Hiroyuki Terashima, Michio Homma (*Div. Biol. Sci., Grad. Sch. Sci., Nagoya University*)

1Pos155 ケラトサイトの回転するストレスファイバ車輪の内側の核と外側の細胞膜の動き
Movement of inner nucleus and outer cell membrane of a rotating stress fiber-wheel in a migrating keratocyte
Chika Okimura, Yoshiaki Iwadate (*Fac. Sci., Yamaguchi Univ.*)

1Pos156 バクテリアべん毛タンパク質輸送装置のゲート機構
Gating mechanism of the bacterial flagellar protein export apparatus
Miki Kinoshita[1], Keiichi Namba[1,2,3], Tohru Minamino[1] ([1]*Grad. Sch. Frontier Biosci, Osaka Univ.*, [2]*RIKEN, Spring-8*, [3]*RIKEN, BDR*)

1Pos157 スピロヘータの遊泳におけるべん毛回転と細胞形状の関係
Relationship between the flagellar rotation and cell shape in a swimming spirochete
Toshiki Kuribayashi, Shuichi Nakamura (*Grad. Sch. Eng., Tohoku Univ.*)

1Pos158 正のべん毛本数制御因子 FlhF は MS リング構成因子 FliF の極局在を促進する
The polar localization of FliF, composing MS-ring, is promoted by FlhF in Vibrio alginolyticus
Yuna Inoue, Seiji Kojima, Keiichi Hirano, Hiroyuki Terashima, Michio Homma (*Division of Biological Science, Graduate School of Science, Nagoya University*)

1Pos159 超解像顕微鏡法と単粒子追跡法による標的細胞へのエクソソーム取り込み機構の解明
Uptake mechanism of exosomes by target cells as revealed by super-resolution microscopy and single-particle tracking
Koichiro M. Hirosawa[1], Taka A. Tsunoyama[2], Yasunari Yokota[3], Kenichi G. N. Suzuki[1,4] ([1]*G-CHAIN, Gifu Univ.*, [2]*OIST*, [3]*Information Science, Gifu University*, [4]*JST-CREST*)

1Pos160 ミトコンドリアの形態と活性の間に相関はあるか？
Are there any correlations between the morphology and the activity of mitochondria?
Arima Okutani (*Tokyo University of Agriculture and Technology*)

1Pos161 酸素への電子伝達阻害時のミトコンドリアの分極機構
Mitochondrial polarization upon inhibition of electron transfer to oxygen
Hinako Tanaka[1], Emika Shida[1], Ikuroh Ohsawa[2], Yoshihiro Ohta[1] ([1]*Grad. Sch. Biotech., TUAT*, [2]*Bioregulatory function., TMGH*)

1Pos162 Single-molecule imaging of PI3K in eukaryotic motile cell
Satomi Matsuoka[1,2,3], Seiya Fukushima[1], Masahiro Ueda[1,2] ([1]*Graduate School of Frontier Biosciences, Osaka University*, [2]*Center for Biosystems Dynamics Research, RIKEN*, [3]*PRESTO, JST*)

1Pos163 温度条件に依存して変化する、大腸菌走化性適応過程の定量解析
Quantitative analysis of E. coli chemotaxis adaptation process that changes depending on temperature conditions
Hiroto Tanaka, Yasuaki Kazuta, Hiroaki Kojima (*Adv ICT Res Inst, NICT*)

1Pos164 細胞辺縁での Aβ の凝集は細胞運動を抑制した
Aggregation of Aβ at the cell periphery suppressed cell motility
Yusaku Chikai, Ryota Yamashita, Masahiro Kuragano, Kiyotaka Tokuraku (*Grad. Sch. Eng., Muroran Inst. Tech.*)

1Pos165 Mechanisms of negative gravitaxis in free-swimming *Chlamydomonas reinhardtii*
Azusa Kage[1], Toshihiro Omori[2] ([1]*Dept. Mech. Eng., Toyohashi U. Tech.*, [2]*Dept. Finemechanics, Tohoku U.*)

1Pos166 孤立した MDCK 細胞における形態および運動の多様性
Diversity in morphological and motile patterns of isolated MDCK epithelial cells
Shimon Shibagaki[1], Shota Mise[1], Seiya Nishikawa[1], Hiroko Nakamura[2], Hiroshi Kimura[2], Atsuko Takamatsu[1] ([1]*Dept. of Elec., Eng. & Biosci., Waseda Univ.*, [2]*Dept. of Mecha., Tokai Univ.*)

生体膜・人工膜／Biological & Artificial membrane: Structure & Property

1Pos167* リン脂質スクランブラーゼ XKR による昆虫細胞膜の高粘弾性変形能
Enhanced viscoelastic deformation of insect cell membrane by phospholipid scramblase XKR
Akifumi Shiomi[1], Kohjiro Nagao[1], Akihisa Yamamoto[2], Ryo Suzuki[2], Motomu Tanaka[2], Masato Umeda[1] ([1]*Grad. Sch. Eng., Kyoto Univ.*, [2]*Inst. Adv. Stud., Kyoto Univ.*)

1Pos168* ピコ秒時間分解けい光分光法で評価した膜標的薬物の人工脂質二重膜への影響
Effect of membrane-targeted drugs on artificial lipid bilayer membranes evaluated by picosecond time-resolved fluorescence spectroscopy
Natsumi Okada[1], Masayuki Iwamoto[2], Haruna Hayashi[1], Akira Takakado[1], Shigetoshi Oiki[3], Koichi Iwata[1] ([1]*Faculty of Science, Gakushuin University*, [2]*Faculty of Medical Sciences, University of Fukui*, [3]*Biomedical Imaging Research Center, University of Fukui*)

1Pos169* ボトムアップ配列設計による α-ヘリックスペプチドナノポアの構築
De novo design for pore-forming peptides with α-helical structure
Masataka Usami, Keisuke Shimizu, Yusuke Sekiya, Ryuji Kawano (*Tokyo University of Agriculture and Technology*)

1Pos170* 脂質膜中にナノポアを構築する β シートペプチドの De novo 配列設計
De novo design of nanopore-forming β-sheet peptide in bilayer lipid membrane
Keisuke Shimizu[1], Shungo Sakashita[2], Yoshio Hamada[2], Kenji Usui[2], Batsaikhan Mijiddorj[3], Izuru Kawamura[1], Ryuji Kawano[1] ([1]*Department of Biotechnology and Life Science, Tokyo University of Agriculture and Technology*, [2]*Faculty of Frontiers of Innovative Research in Science and Technology, Konan University*, [3]*Department of Materials Science and Engineering, Yokohama National University*)

1Pos171* Investigation of local anesthetic and membrane interactions using model cell membranes
Wanjae Choi[1], Hyunil Ryu[1], Seulmini Goh[1], Chaoge Zhou[1], Soonjo Kwon[1], Sun Min Kim[2], Tae-Joon Jeon[1] ([1]*Department of Biological Engineering, Inha University, Incheon*, [2]*Department of Mechanical Engineering, Inha University, Incheon*)

1Pos172 環状ジペプチドの安定構造における系統的な傾向と特徴
Systematic trends and features in the stable structure of cyclic dipeptides
Koki Yanagi[1,2], Hiroaki Nishizawa[2], Ryunosuke Yoshino[2], Ryuhei Harada[2], Yasuteru Shigeta[2] ([1]*Phys., Pure and Applied Sci., Grad. Sch. Univ. Tsukuba*, [2]*CCS, Univ. Tsukuba*)

1Pos173 Ca2+-dependent high-conductance channel activity of F-ATP synthase matches the permeability transition pore
Andrea Urbani[1,2,5,6], Valentina Giorgio[2], Andrea Carrer[2], Cinzia Franchin[2], Giorgio Arrigoni[2], Chimari Jiko[3], Kazuhiro Abe[7], Janna F.M. Bogers[4], Shintaro Maeda[6], Kyoko Shinzawa[5], **Christoph Gerle**[1], Ildiko Szabo[2], Paolo Bernardi[2] ([1]*IPR*, [2]*Univ. Padova*, [3]*Kyoto Univ.*, [4]*TU Delft*, [5]*Univ. Hyogo*, [6]*Scripps*, [7]*Nagoya Univ.*)

1Pos174 P-SPICA: A coarse-grained force field for biological systems with a polar coarse-grained water model
Yusuke Miyazaki, Susumu Okazaki, Wataru Shinoda (*Grad. Sch. Eng., Nagoya Univ.*)

1Pos175 光照射によるリポソーム内への配列選択的 DNA 輸送
Sequence selective DNA transport into liposome by photoirradiation
Shigetaka Nakamura, Nobuharu Uehara, Takashi Hasegawa, Kenzo Fujimoto (*JAIST*)

1Pos176 生細胞膜上における相分離の誘発と可逆的制御
Induction and reversible control of phase separation on living cell membranes
Kenichi Kawano[1], Yasufumi Takahashi[2], Ryo Ohtani[3], Masanao Kinoshita[4], Shiroh Futaki[1] ([1]*Institute for Chemical Research, Kyoto University*, [2]*WPI-NanoLSI, Kanazawa University*, [3]*Department of Chemistry, Faculty of Science, Kyushu University*, [4]*Graduate School of Science, Kyushu University*)

1Pos177 気液界面における人工肺サーファクタント膜への微粒子の影響
Effect of sub-micron particles on a model lung surfactant monolayer at the air-water interface
Masahiro Hibino[1], Toshiki Kamata[2] ([1]*Div. Sust. Enviro. Eng., Muroran Inst. Tech.*, [2]*Dept. Appl. Sci., Muroran Inst. Tech.*)

1Pos178 On the condensing effects of 7β-hydroxycholesterol, 25-hydroxycholesterol, and cholesterol on DPPC bilayers
Hiroshi Takahashi, Tatsuya Hoshino (*Biophys. Lab., Grad. Sch. Sci.&Tech., Gunma Univ.*)

1Pos179 蛍光寿命測定を用いた脂質二重膜におけるセラミド-1-リン酸の動的挙動解析
Dynamic behavior of ceramide-1-phosphate in lipid bilayers examined by fluorescence lifetime measurement
Tomokazu Yasuda[1,2], J. Peter Slotte[3] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Research Foundation Itsuu Laboratory*, [3]*Åbo Akademi Univ.*)

1Pos180 Molecular dynamics simulation of the mechanical properties of lipid membranes in the presence of proteins
Diego Ugarte, Shoji Takada (*Takada Lab., Dept. Biophysics, Div. Biology, Grad. Sch. of Sci., Kyoto Univ.*)

1Pos181 EGFR 膜近傍領域のリン酸化が EGFR TM-JM 二量体構造に及ぼす影響
The impact of phosphorylation in the EGFR JM region on the dimer structure of EGFR TM-JM region
Daisuke Matsuoka[1], Yuji Sugita[1,2,3] ([1]*RIKEN Theoretical Molecular Science*, [2]*RIKEN R-CCS*, [3]*RIKEN BDR*)

1Pos182 The dipole potential probed by hydrophobic ions using the contact bubble bilayer method
Yuka Matsuki[1], Masayuki Iwamoto[2], Mariko Yamatake[2], Shigetoshi Oiki[3] ([1]*Dept. Anesth & Reanmat., Univ. Fukui Facult. Med. Sci.*, [2]*Dept. Mol. Neurosci., Univ. Fukui Facult. Med. Sci.*, [3]*Biomed. Imaging Res. Center, Univ. Fukui*)

1Pos183 膜活性な両親媒性ランダムコポリマーによる脂質ナノディスクの自発形成
Spontaneous formation of lipid nanodisc by membrane active amphiphilic random copolymer
Kazuma Yasuhara, Mitsuyoshi Yuma, Jinyu Hao, Jin Arakida, Rapenne Gwenael, Jun-ichi Kikuchi (*Div. Mater. Sci., Nara Inst. Sci. Tech.*)

1Pos184 動的および静的光散乱法によるリン脂質ベシクルの構造評価 2
Structural evaluation of phospholipid vesicles by dynamic and static light scattering techniques 2
Nobutake Tamai[1], Takeshi Nobuoka[2], Ryo Takechi[2], Masaki Goto[1], Hitoshi Matsuki[1] ([1]*Grad. Sch. Tech. Indus. Soc. Sci., Tokushima Univ.*, [2]*Dept. Biol. Tech. Sci., Col. Eng., Tokushima Univ.*)

1Pos185 脂質膜外葉のみに形成したラフト様秩序領域の物性とそれが内葉に及ぼす影響
Physicochemical properties of raft-like ordered domains formed in outer leaflet and its influence on the inner leaflet
Takuya Koga, Masanao Kinoshita, Nobuaki Matsumori (*Grad. Sch. Sci., Kyushu Univ.*)

1Pos186 三種のリン脂質/コレステロール・リポソームにおけるクロルゾキサゾン捕捉量とコレステロール濃度との相関
Correlation between the amount of trap of chlorzoxazone by various phospholipid/cholesterol liposomes and their cholesterol concentrations
Shosei Kano, Hiroshi Takahashi (*Biophys. Lab., Grad. Sch. Sci.&Tech., Gunma Univ.*)

1Pos187 ジミリストイルホスファチジルコリン 2 重膜に形成された脂質様錯体ドメインの物理化学的特性
Physico-chemical properties of lipophilic complex-rich domains formed in dimyistoylphosphatidylcholine (DMPC) bilayers
Hikaru Watanabe, Yoshinao Kinoshita, Ryo Ohtani, Nobuaki Matsumori (*Grad. Sch. Sci., Kyushu Univ.*)

1Pos188 低流量電子線散乱法を用いた局所的な脂質充填構造の解析
Low-flux election-diffraction discloses the local structure of lipid membrane
Shimpei Yamaguchi, Masanao Kinoshita, Nobuaki Matsumori (*Grad. Sch. Sci., Kyushu Univ.*)

化学受容／Chemoreception

1Pos189* 全反射赤外分光法によるムスカリン性アセチルコリン M2 受容体のリガンド認識、活性化機構研究
Ligand recognition and activation mechanism in muscarinic acetylcholine receptor M2 (M2R) study by ATR-FTIR spectroscopy
Kohei Suzuki[1], Kodai Katayama[1], Ryoji Suno[2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Kansai Medical University*)

1Pos190 コレラ菌タウリン走性受容体 Mlp37 の制御ネットワーク
The regulatory network controls expression of the taurine chemoreceptor Mlp37 in *Vibrio cholerae*
So-ichiro Nishiyama[1], Hirotaka Tajima[2,3], Shiori Onogi[2], Hiroshi Urakami[1], Ikuro Kawagishi[2,3] ([1]*Fac. App. Life Sci., Niigata Univ. Pharm. App. Life Sci.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

1Pos191 大腸菌グローバル転写因子 PdhR による走化性受容体遺伝子発現調節
Expression of the major chemoreceptor genes is regulated by a global transcription factor PdhR in *Escherichia coli*
Ayano Inoue[1], Nana Ito[1], Yumeno Kawasaki[1], Eri Shiokawa[1], Hirotaka Tajima[2,3], Ikuro Kawagishi[1,2,3] ([1]*Grad. Sch.Sci.&Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

1Pos192 大腸菌細胞側面膜領域における走化性受容体クラスター形成
Chemoreceptor clustering of *Escherichia coli* in lateral regions of the cytoplasmic membrane
Nana Ito[1], Masatoshi Nishikawa[2], Yoshiyuki Sowa[1,2,3], Ikuro Kawagishi[1,2,3] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

1Pos193 細菌の可溶性走化性受容体と細胞膜貫通型走化性トランスデューサー相互作用解析
Probing interaction between a soluble receptor and a transmembrane transducer in bacterial chemotaxis
Hisashi Kubota[1], Kana Murakami[2], Hirotaka Tajima[2,3], Ikuro Kawagishi[1,2,3] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

神経・感覚／Neuroscience & Sensory systems

1Pos194* 脳脊髄液流動を想定した非平衡空間におけるアミロイド β 凝集
Amyloid β Aggregation in Non-equilibrium Space Mimicking Cerebrospinal Fluidic Flow
Akane Iida[1], Kei Unoura[2], Hideki Nabika[2] ([1]*Graduate School of Sci. and Eng., Yamagata Univ.*, [2]*Faculty of Sci., Yamagata Univ.*)

1Pos195* 光ピンセットを用いた AMPA 型グルタミン酸受容体分子操作における神経電気活動変化
Neuronal electrical activity induced by optical trapping of AMPA-type glutamate receptors on neurons
Tatsunori Kishimoto[1,2], Suguru Kudoh[2], Takahisa Taguchi[3], Chie Hosokawa[1,2,4] ([1]*Grad. Sch. Sci., Univ. Osaka City*, [2]*Grad. Sch. Sci. Tech., Univ. Kwansei Gakuin*, [3]*NICT,CiNet*, [4]*PhotoBIO-OIL, AIST-Osaka Univ.*)

1Pos196 (1SEP-2) Single-cell trajectory analysis of human iPS cell-derived neurons carrying a rare RELN deletion
Yuko Arioka[1,2,3], Emiko Shishido[1,4], Norio Ozaki[1] ([1]*Department of Psychiatry, Nagoya University Graduate School of Medicine*, [2]*Nagoya University Hospital*, [3]*Institute for Advanced Research, Nagoya University*, [4]*National Institute for Physiological Sciences*)

1Pos197 ミミズの短期記憶メカニズム
Molecular mechanism of short-term memory formation in earthworms
Yoshiichiro Kitamura, Akira Sakane, Hikaru Tsumita (*Department of Mathematical Sciences and Physics, College of Science and Engineering, Kanto Gakuin University*)

神経回路・脳の情報処理／Neuronal circuit & Information processing

1Pos198* 時間依存性の環境変化認識のモデリング：光結合する矩形波発振子のネットワークがもたらす幾何学的フラストレーション
Perception of time-dependent environmental change: A toy model with photo-coupled electronic oscillators composing frustrated network
Hiroshi Ueno, Masatomo Matsushima (*Dep. Med. Info., Grad. Sch. Life Med. Sci., Doshisha Univ.*)

1Pos199 大脳皮質神経細胞の単一配置による神経回路の構築
Construction of neural network with arrangement of single cerebral cortical neuron
Hayato Toriumi, Tomoyuki Kaneko (*LaRC,FB,Hosei Univ*)

1Pos200 緑茶由来カテキンはナメクジの匂い嫌悪条件付け学習による記憶形成を強化する
Green tea-derived catechins enhance the long-term memory formation for an odor-aversive conditioning in the land slug
Yoshimasa Komatsuzaki[1], Keisuke Matsui[2], Kyouka Ishizuka[1], Kouki Tezuka[1], Ken Lukowiak[3], Minoru Saito[4] ([1]*CST, Nihon Univ.*, [2]*Grad. Sch. of Sci. and Tech., Nihon Univ.*, [3]*Hotchkiss Brain Inst., Fac. Med., Univ. Calgary*, [4]*Dept. Biosci., Coll. Hum. Sci., Nihon Univ.*)

1Pos201 Sites for formation and storage of associative motor memory revealed by reversible expression of metabotropic glutamate receptor 1
Yasushi Kishimoto[1], Harumi Nakao[2], Kouichi Hashimoto[3], Kazuo Kitamura[4], Miwako Yamasaki[5], Kazuki Nakao[2], Masahiko Watanabe[5], Masanobu Kano[6], Atsu Aiba[2], Yutaka Kirino[1] ([1]*Kagawa. Sch. Pharm. Sci., Tokushima Bunri Univ.*, [2]*Cent. Dis. Biol. Integr. Med., Univ. Tokyo*, [3]*Sch. Med., Hiroshima Univ.*, [4]*Sch. Med., Univ. Yamanashi*, [5]*Sch. Med., Hokkaido Univ.*, [6]*Sch. Med., Univ. Tokyo*)

光生物学：視覚・光受容／Photobiology: Vision & Photoreception

1Pos202* 光センサー LOV2 ドメインの光反応中間体の構造揺らぎ検出
Time-resolved study on structural fluctuations of transient intermediates of the light sensor LOV2 domain
Shunrou Tokonami, Yusuke Nakasone, Masahide Terazima (*Dep. Chem., Sch. Sci., Kyoto Univ.*)

1Pos203* LOV ドメイン型光活性化アデニル酸シクラーゼ mPAC の光反応ダイナミクス
Photoreaction dynamics of LOV-domain-regulated photoactivated adenylate cyclase mPAC
Misato Ikoma, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci., Univ. Kyoto*)

1Pos204* 光センサータンパク質 PYP と下流分子 PBP による励起波長依存的な会合・解離反応ダイナミクス
Excitation wavelength-dependent association and dissociation dynamics between light sensor protein PYP and its downstream protein PBP
Suhyang Kim[1], Yusuke Nakasone[1], Akira Takakado[2], Yoichi Yamazaki[3], Hironari Kamikubo[3], Masahide Terazima[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*Grad. Sch. Sci., Univ. Gakushuin*, [3]*Div. Mat. Sci., NAIST*)

1Pos205* TG 法を用いた OCP と FRP の時間分解相互作用ダイナミクスの検出
Detection of time-resolved interaction between OCP and FRP by using transient grating method
Takatoshi Ohata, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci., Univ. Kyoto*)

1Pos206* 固体 NMR による KR2 のレチナール結合ポケットと Na+結合サイト間の水素結合ネットワークの構造解析
Structural analysis of hydrogen-bond networks between retinal binding pocket and Na+ binding site on KR2 by solid-state NMR
Rina Kaneko[1], Arisu Shigeta[2], Toshio Nagashima[3], Toshio Yamazaki[3], Keiichi Inoue[4,5], Hideki Kandori[5], Izuru Kawamura[1,2] ([1]*Grad. Sch. Eng. Sci., Yokohama National Univ.*, [2]*Grad. Sch. Eng., Yokohama National Univ.*, [3]*RIKEN*, [4]*Univ. Tokyo*, [5]*Nagoya Inst. Tech.*)

1Pos207 SEC-SAXS 法によるシロイヌナズナ由来フィトクロム B の構造解析
Structural analysis of Arabidopsis phytochrome B by small-angle X-ray scattering coupled with size-exclusion chromatography
Mao Oide[1,2], Takaaki Hikima[2], Tomotaka Oroguchi[1,2], Takayuki Kato[3], Yuki Yamaguchi[1,2], Shizue Yoshihara[4], Masaki Yamamoto[2], Masayoshi Nakasako[1,2], Koji Okajima[1,2] ([1]*Grad. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 center*, [3]*Grad. Sci. of Front. Biosci., Osaka Univ.*, [4]*Dept. of Biol. Sci., Osaka Pref. Univ.*)

1Pos208 Structural basis of photo-stability of invertebrate rhodopsins
Midori Murakami (*Dept Physics, Nagoya Univ*)

1Pos209 ロドプシンの構成的活性変異体 M257Y のメカニズムに関する分子動力学シミュレーション
Molecular dynamics simulation study on the mechanism of constitutively active mutant M257Y of rhodopsin
Yuichiro Kanamori, Tadaomi Furuta, **Minoru Sakurai** (*Tokyo Tech*)

1Pos210 ラマン光学活性で観るハロロドプシン多量体形成による活性部位の構造変化
Raman optical activity observes a clear structural change of active site caused by trimer formation of halorhodopsin
Shogo Ogawa[1], Tomotsumi Fujisawa[1], Takashi Kikukawa[2,3], Masashi Unno[1] ([1]*Grad. Sch. Sci. Eng., Saga Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*GSS, GI-CoRE, Hokkaido Univ.*)

1Pos211 ラマン光学活性による光駆動型内向きプロトンポンプの研究
Near-IR Raman optical activity spectroscopy of inward proton pump rhodopsin
Ryosuke Kuroiwa[1], Tomotsumi Fujusawa[2], Yuki Sudo[3,4], Megumi Kamimura[2], Saki Inoue[3], Masashi Unno[2] ([1]*Grad. Sch. Sci. Eng., Saga Univ.*, [2]*Fac. Sci. Eng., Saga Univ.*, [3]*Grad. Sch. of Med. Dent. & Pharm. Sci., Okayama Univ.*, [4]*Fac. of Pharm. Sci., Okayama Univ.*)

1Pos212 Actinotalea fementans 由来ヘリオロドプシンの物性解析
Molecular properties of Heliorhodopsin from Actinotalea fementans
Rei Abe-Yoshizumi, Ai Muto, Hideki Kandori (*Nagoya Inst. Tech.*)

1Pos213 (6-4)光回復酵素の光反応過程における時間分解分光研究
Time-resolved spectroscopic study on photoreaction of (6-4) photolyase
Daichi Yamada[1], Takashi Nomura[1], Yuna Nakajima[2], Minoru Kubo[1] ([1]*Grad. Sch. Life Sci.*, [2]*Dept. Life Sci., Univ. Hyogo, Japan*)

1Pos214 集光クマリン色素を有する DNA 光回復酵素による高効率光駆動 DNA 修復
Enhanced light-driven DNA repair by DNA photolyase bearing light-harvesting coumarin chromophore
Yuma Terai[1], Ryuma Sato[2], Risa Matsumura[1], Shigenori Iwai[1], **Junpei Yamamoto**[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*RIKEN BDR*)

1Pos215 Impact of a water molecule on photoreduction of (6-4) photolyase
Yuhei Hosokawa[1], Ryuma Sato[2], Shigenori Iwai[1], Junpei Yamamoto[1] ([1]*Grad. Sch. Eng. Sci., Univ. Osaka*, [2]*Riken*)

1Pos216 Characterization of Antarctic inward proton pumping microbial rhodopsins (AntRs)
Andrew Harris[1], Mizuho Tomita[2], Luiz Schubert[3], Michalis Lazaratos[4], Ethan Watt[1], Anh Hoang[1], Ana-Nicoleta Bondar[4], Joachim Heberle[3], Furutani Yuji[2], Hideki Kandori[2], Leonid Brown[1] ([1]*University of Guelph, Physics*, [2]*Nagoya Institute of Technology, Life Science and Applied Chemistry*, [3]*Freie Universitat Berlin, Experimental Physics*, [4]*Freie Universitat Berlin, Theoretical Physics*)

1Pos217 Inversion of Proton Transport Direction in Thermophilic Rhodopsin by Neutralizing the Secondary Counterion Asp229
Minori Kiyoshima[1], Takashi Kikukawa[2,3], Tomoyasu Aizawa[2,3], Makoto Demura[2,3], Yuki Sudo[4], Takashi Tsukamoto[2,3] ([1]*Grad. Sch. Life Sci. Hokkaido Univ.*, [2]*Fac. Adv. Life Sci. Hokkaido Univ.*, [3]*GSS, GI-CoRE*, [4]*Grad. Sch. Med. Dent. Pharm. Sci. Okayama Univ.*)

1Pos218 シゾロドプシンの内向きプロトン輸送経路の特性
Characterization of the inward proton transport pathway in Schizorhodopsin
Masae Konno[1,2], Keiichi Inoue[1,3], Rohit Ghai[4], Oded Beja[5], Hideki Kandori[1,2] ([1]*Life Sci. Appl. Chem., Grad. Sch. Eng., NIT*, [2]*OBTRC, NIT*, [3]*ISSP, Univ. Tokyo*, [4]*Aquatic Microbial Ecology, Biology Centre CAS, Inst. Hydrobiol.*, [5]*Technion - Israel Inst. Tech.*)

光生物学：光合成／Photobiology: Photosynthesis

1Pos219* 紅色光合成細菌由来光捕集反応中心 1 複合体のスペクトル多様性と安定性におけるカルシウムイオンの役割
A dual role for calcium in expanding the spectral diversity and stability of LH1-RC photocomplexes of purple phototrophic bacteria
Michie Imanishi[1], Mizuki Takenouchi[2], Shinichi Takaichi[3], Shiori Nakagawa[4], Yoshitaka Saga[4], Shinji Takenaka[1], Michael Madigan. T[5], Jorg Overmann[6], Zheng-Yu Wang-Otomo[2], Yukihiro Kimura[1] ([1]*Grad. Sch. Agricultural Sci., Kobe Univ.*, [2]*Fac. of Sci., Ibaraki Univ.*, [3]*Fac. of Life Sci., Tokyo Univ. of Agriculture*, [4]*Dep. of Chem., Kindai Univ.*, [5]*Dep. of Microbiol.., Southern Illinois Univ.*, [6]*Microbiol., Braunschweig Univ. of Tech.*)

1Pos220 超分子複合体構造を構成する光合成アンテナ組成の解明
Elucidation of supramolecular components in photosynthetic antenna
Tetsuko Nakaniwa[1], Ryuichi Kano[2], Naoko Norioka[1], Soichiro Seki[2], Ritsuko Fujii[2,3], Genji Kurisu[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka City Univ.*, [3]*OCARINA, Osaka City Univ.*)

1Pos221 Molecular mechanism of pH-dependent electron-flow regulation in photosystem II
Yuichiro Shimada[1], Seiryu Nakajima[1], Ryo Nagao[1,2], Takumi Noguchi[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*RIIS, Okayama Univ.*)

1Pos222 緑藻の光捕集アンテナタンパク質 SCP の再構成
In-vitro reconstitution of light-harvesting complexes of a siphonous green alga, *Codium fragile*
Yuki Isaji[1], Nami Yamano[1,2], Masahiko Iha[3], Tetsuko Nakaniwa[4], Rei Toda[5], Naoko Norioka[4], Genji Kurisu[4,5], Ritsuko Fujii[1,2] ([1]*Grad. Sch. Sci., Osaka City Univ*, [2]*OCARINA, Osaka City Univ.*, [3]*SouthProduct Co. Ltd.*, [4]*IPR, Osaka Univ.*, [5]*Grad. Sch. Sci., Osaka Univ.*)

1Pos223 LH2 タンパク質の B800 部位へ再構成したクロロフィル誘導体のスペクトル特性変化に対するテトラピロール環構造の影響
Structural effects of chlorophyll pigments on their spectral properties induced by reconstitution into the B800 site in LH2 protein
Yoshitaka Saga, Madoka Yamashita, Kanji Miyagi (*Faculty of Science and Engineering, Kindai University*)

1Pos224 ガラス基板表面におけるチラコイド膜の再構成と光合成機能解析
Reconstitution and functional analysis of thylakoid membrane on a glass substrate
Takuro Yoneda[1], Yasushi Tanimoto[1], Daisuke Takagi[2], Kenichi Morigaki[1,3] ([1]*Grad. Sch. Agr., Univ. Kobe*, [2]*Grad. Sch. Agr., Univ. Tohoku*, [3]*Biosignal., Univ. Kobe*)

1Pos225 光化学系複合体と酸化グラフェンを用いた水素発生
Hydrogen production using photosystem and graphene oxide
Shunsuke Sone[1], Mriko Miyachi[2], Shota Tanaka[1], Hisataka Ohta[1], Yoshinori Yamanoi[2], Akihide Iwase[1], Akihiko Kudo[1], Hiroshi Nishihara[2], Tatsuya Tomo[1] ([1]*Tokyo University of science*, [2]*The University of Tokyo*)

1Pos226 緑藻ミル糸状体のカロテノイド蓄積における培養時光条件の影響
The effect of different light regimes for carotenoid accumulation of a macro green algae, Codium fragile, in filamentous form
Soichiro Seki (*Osaka city university , department of Chemistry, Research institute for natural science and technology*)

1Pos227 Time-resolved infrared analysis of proton release pathways in photosynthetic water oxidation using a D1-N298A mutant and NO_3^- substitution
Yasutada Okamoto[1], Yuichiro Shimada[1], Ryo Nagao[1,2], Takumi Noguchi[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*RIIS, Okayama Univ.*)

1Pos228 QM/MM analysis of the protonation structure of the S_0 state in the water-oxidizing Mn_4CaO_5 cluster
Masao Yamamoto, Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

1Pos229 Infrared microspectroscopic study on water oxidation in a single photosystem II microcrystal
Yuki Kato[1], Satoshi Haniu[1], Yoshiki Nakajima[2], Fusamichi Akita[2,3], Jian-Ren Shen[2], Takumi Noguchi[1] ([1]*Grad. Sch. Sci, Nagoya Univ.*, [2]*Res. Inst. Interdiscip. Sci., Okayama Univ.*, [3]*JST-PRESTO*)

1Pos230 光化学系 II の表在性タンパク質による水分解 Mn_4CaO_5 クラスターの S_2 構造異性体平衡の制御機構
Equilibrium of the S_2-state isomers of the water-oxidizing Mn_4CaO_5 cluster in photosystem II regulated by extrinsic proteins
Shota Taguchi[1], Liangliang Shen[2], Guangye Han[2], Jian-Ren Shen[3], Takumi Noguchi[1], Hiroyuki Mino[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Key Lab. Photobiol., Inst. Botany, Chinese Acad. Sci., China*, [3]*Res. Inst. Interdiscip. Sci., Okayama Univ.*)

1Pos231 Dynamics of photosystem II protein complexes as observed by high speed atomic force microscopy
Takaya Tokano[1], Yuki Kato[1], Shogo Sugiyama[2], Takumi Noguchi[1], Takayuki Uchihashi[1,3] ([1]*Grad.Sch.Sci.,Nagoya Univ.*, [2]*Grad.Sch.Phys.,Kanazawa Univ*, [3]*EXCELLS*)

1Pos232* 酵素型ロドプシン（Rh—PDE）の非対称的 pH 効果
Asymmetric pH effect on the enzyme rhodopsin, Rh-PDE
Masahiro Sugiura[1], Kazuho Yoshida[1], Masahiro Hibi[3], Satoshi Tsunoda[1,2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*JST PRESTO*, [3]*Graduate School of Science, Nagoya University*)

1Pos233* 光活性型 bZIP モジュールであるフォトジッパーにおける Gln317 の役割
The role of Gln317 in a light-activatable bZIP module, Photozipper
Itsuki Kobayashi, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

1Pos234* プロトンポンプ型ロドプシンによる緑藻クラミドモナスの 非光化学的消光(NPQ)の人為的光制御
Optical control of non-photochemical quenching (NPQ) in the alga *Chlamydomonas reinhardtii* by light-driven proton pump rhodopsins
Yurie Nagase[1], Keiichi Kojima[1], Saki Inoue[1], Hiroshi Kuroda[2], Ryutaro Tokutsu[3], Shinji Masuda[4], Jun Minagawa[3], Yuichiro Takahashi[2], Yuki Sudo[1] ([1]*Grad. Sch. of Med. Dent. & Pharm. Sci. Okayama Univ.*, [2]*RIIS., Okayama Univ.*, [3]*Div. of Environ. Photobiol., NIBB*, [4]*Cent. Biolog. Resources & Informatics, Tokyo Inst. Technol.*)

1Pos235* Theoretical study on molecular mechanics of natural anion channelrhodopsin GtACR1
Takafumi Shikakura, Cheng Cheng, Shigehiko Hayashi (*Kyoto Univ.*)

生命の起源・進化／Origin of life & Evolution

1Pos236 **蝶のカモフラージュや擬態模様にみる多要素構造**
Multi-component systems of camouflage and mimicry in butterfly wing patterns
Takao Suzuki (*NARO*)

1Pos237 **巨大化大腸菌の再生過程可視化**
Regeneration of Escherichia coli giant protoplasts
Kazuhito Tabata, Takao Sogo, Yoshiki Moriizumi, Hiroyuki Noji (*Department of Applied Chemistry, The University of Tokyo*)

ゲノム生物学／Genome biology

1Pos238 **大腸菌を用いた実験室内進化におけるタンパク質の配列進化速度の制約**
Constraint of protein evolution speed in de novo experimental evolution of *E. coli*
Saburo Tsuru[1], Atsushi Shibai[2], Chikara Furusawa[2] ([1]*Sch. of Sci, The Univ. of Tokyo*, [2]*RIKEN BDR*)

1Pos239 (1SEA-5) Molecular Dynamics of Nucleosome Assembly
Giovanni Brandani, Shoji Takada, Cheng Tan (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

生命情報科学／Bioinformatics

1Pos240 **全原子 Motion Tree による側鎖運動の記述とドメイン運動との連動**
Full-atom Motion Tree detects side-chain motions and their coupling with domain motions
Ryotaro Koike, Motonori Ota (*Grad. Sch. Info., Nagoya Univ.*)

1Pos241 **マルチカノニカル法を用いた蛋白質球状ドメイン外の相互作用の解析**
Analysis of the protein-protein interaction between regions external to globular domains with multi-canonical molecular method
Takuya Shimato[1], Takuya Takahashi[2], Kota Kasahara[2], Junichi Higo[3] ([1]*Grad. Sch. Life Sci., Ritsumeikan Univ.*, [2]*Coll. Life Sci., Ritsumeikan Univ.*, [3]*Grad. Sch. Sim. Studies, Univ. Hyogo*)

1Pos242 **並列タンパク質間相互作用予測システム MEGADOCK の高速化・仮想化**
Acceleration and virtualization of parallel protein-protein interaction prediction system MEGADOCK
Masahito Ohue[1], Hiroki Watanabe[1,2], Kento Aoyama[1,2], Yutaka Akiyama[1] ([1]*Sch Computing, Tokyo Tech*, [2]*RWBC-OIL, AIST*)

1Pos243 **蛋白質における Non-local 接触を持たない領域に関する統計解析**
Segments without non-local contacts in protein structures
Kota Kasahara[1], Shintaro Minami[2], Yasunori Aizawa[3], Ryohei Kondo[4], Takuya Shimato[4], Takuya Takahashi[1] ([1]*Coll. Life. Sci., Ristumeikan Univ.*, [2]*EXCELLS, NINS*, [3]*Sch. Life Sci., Tech., TokyoTech*, [4]*Grad. Sch. Life Sci., Ritsumeikan Univ.*)

1Pos244 Direct coupling analysis of amino-acid sequences based on the Hopfield-Potts model
Kai Shimagaki, Martin Weigt (*Sorbonne Universite, Paris-IV*)

数理生物学／Mathematical biology

1Pos245 **繊維状粒子凝集の CA タイプ解析**
Attempts at CA-type formal analysis of fibrous assembly of particles
Takashi Konno (*Biomath.Med.Univ.Fukui*)

1Pos246 エピジェネティックな状態変化が細胞のがん化に及ぼす影響のランドスケープ理論による解析
Landscape analyses of epigenetic state change in cancerization
Yutaro Kameyama, Masaki Sasai (*Dept. Appl. Phys., Grad. Sch. Eng., Univ. Nagoya*)

1Pos247 筋分化過程で誘導される細胞競合は IGF シグナルを同期化する
Synchronization of IGF signal by cell competition during myogenesis
Fumihiko Hakuno, Masato Masuda, Ryosuke Okino, Shin-Ichiro Takahashi (*Dep. App. Ani. Sci., Grad. Sch. Agri. Life. Sci., The Univ. of Tokyo.*)

1Pos248 モジホコリ変形体における輸送管ネットワークの分岐則
Direct observation of branching rules in transportation network of *Physarum* plasmodium
Masahiro Shibata, Atsuko Takamatsu (*Dept. of Elec., Eng. & Biosci., Waseda Univ.*)

1Pos249 シグナル伝達分子の細胞膜上クラスター形成機構の数理研究
Mathematical study on cluster formation of signaling proteins on the cell membrane
Hiroaki Takagi (*Dep. Phys., Sch. Med., Nara Med. Univ.*)

1Pos250 線虫の graded ニューロンはどのようにして確率的な 2 状態スイッチングダイナミクスを生成するか？
How do graded neurons generate stochastic binary switching dynamics in C. elegans?
Yuishi Iwasaki (*Fac. Eng., Ibaraki Univ.*)

非平衡・発生リズム／Nonequilibrium state & Biological rhythm

1Pos251* Effects of in vivo rhythm-damping mutations to KaiA on circadian rhythm in vitro
Masahiro Wakayama[1], Risa Imada[1], Yuki Nakamoto[1], Rie Kumagai[1], Keisuke Serizawa[2], Masahiro Ishiura[3], Kousuke Maki[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Sch. Sci., Nagoya Univ.*, [3]*Nagoya Univ.*)

1Pos252* Exploring a simply phosphorylation cycle by using phosphorylation site variants of clock protein KaiC
Rie Kumagai[1], Risa Imada[1], Shun Terauchi[1], Yuki Nakamoto[1], Masahiro Ishiura[2], Kosuke Maki[1] ([1]*Grad. Sci, Nagoya Univ*, [2]*Nagoya Univ*)

1Pos253 (1SHA-4) 高圧力下で早くなるシアノバクテリアの概日周期
(1SHA-4) Pressure accelerates the circadian clock of cyanobacteria
Ryo Kitahara[1], Katsuaki Oyama[2], Takahiro Kawamura[2], Keita Mitsuhashi[2], Soichiro Kitazawa[1], Kazuhiro Yasunaga[1], Natsuno Sagara[1], Megumi Fujimoto[2], Kazuki Terauchi[2] ([1]*Pharm. Sci., Ritsumeikan Univ.*, [2]*Life Sci., Ritsumeikan Univ.*)

1Pos254 心筋細胞ネットワークにおける局所伝導ゆらぎの幾何学的理解
Geometrical understanding of the local fluctuation in propagation of excitation conduction in cardiomyocyte network
Shota Aoki[1], Kazufumi Sakamoto[1], Yoshitsune Hondo[2], Akihiro Hattori[3], Masao Odaka[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

1Pos255 Observation of direction-dependent asymmetrical propagation velocities in excitation conduction in a same cardiomyocyte networks on a chip
Kazufumi Sakamoto[1], Shota Aoki[1], Yoshitsune Hondo[2], Masao Odaka[3], Akihiro Hattori[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

計測／Measurements

1Pos256 **水素化アモルファスシリコンと紫外可視光変換で増強されたセンサのためのゲル電気化学素子と分子薄膜**
Gel electrochemical element and molecular film for sensor enhanced by hydrogenated amorphous silicon and ultra violet light conversion
Koki Shimanaka[1], Shota Murakami[1], Kairi Shimazaki[1], Kishiro Seino[1], Hikaru Hatakeyama[1], Shu Mugita[1], Hiroshi Masumoto[2], Takashi Goto[3], Yutaka Tsujiuchi[1] ([1]*Mat Sci Akita Univ*, [2]*FRIS Tohoku Univ*, [3]*IMR Tohoku Univ*)

1Pos257 On-chip differential analysis of sequential phagocytosis on identical position of single macrophages
Yuya Furumoto[1], Toshiki Azuma[1], Amane Yoshida[1], Takahiro Kitahara[2], Tomoyasu Sakaguchi[2], Masao Odaka[3], Akihiro Hattori[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

1Pos258 **水素化アモルファスシリコンに積層した脂質とバクテリオロドプシン複合膜の構造変化観察**
Observation of structural change of lipid film and lipid and bacteriorhodopsin film laminated on hydrogenated amorphous silicon film
Hikaru Hatakeyama[1], Hiroshi Masumoto[2], Takashi Goto[3], Yutaka Tsujiuchi[1] ([1]*MatSci AkitaUNIV*, [2]*FRIS TohokuUNIV*, [3]*IMR TohokuUNIV*)

1Pos259 **高速走査レーザーマイクロダイセクションシステムの開発**
Development of a high-speed scanning laser microdissection system
Masahito Hasegawa[1,2], Yasushi Kudo[2], Minako Hirano[1], Hiroaki Yokota[1] ([1]*Grad.Sch.Creation Photon Indust.*, [2]*Disc Tech*)

1Pos260 **光-電子相関顕微鏡法（CLEM）による同一試料観察に向けた相関・位置合わせ精度の評価**
Evaluation of correlation and alignment accuracy toward the same sample observation by CLEM
Yuki Gomibuchi[1], Risa Ezoe[2], Hiroko Takazaki[1], Takuo Yasunaga[1] ([1]*Dept. of Phys. Info. Tech., Kyushu Inst. Tech.*, [2]*Dept. of Biosci. Bioinfo., Kyushu Inst. Tech*)

1Pos261 **原子間力顕微鏡にひと工夫 ー*Volvox* 1 個体の推進力を「直接」測るー**
A trick to atomic force microscopy enabling direct measurement of forces generated by swimming *Volvox* spheroids
Noriyo Mitome[1,2], Kosaku Horinaga[2], Kazumo Wakabayashi[2], Hikaru Emoto[2], Airi Shintome[2], Kazutaka Fujita[3], Noriko Ueki[4], Ken-ichi Wakabayashi[5], **Katsuya Shimabukuro**[2] ([1]*NIT, Chem. Biochem., Numazu Col.*, [2]*NIT, Chem. Bio. Eng., Ube Col.*, [3]*NIT, NIT, Mech., Ube Col.*, [4]*Sci. Res. Cent., Hosei Univ.*, [5]*CLS, Tokyo Tech*)

バイオイメージング／Bioimaging

1Pos262 Intron seqFISH enables transcriptome-wide visualization of genome organization and nascent transcription in single cells
Yodai Takei[1], Sheel Shah[2], Wen Zhou[1], Eric Lubeck[3], Jina Yun[1], Chee-Huat Linus Eng[1], Noushin Koulena[1], Christopher Cronin[1], Christoph Karp[1], Eric Liaw[2], Mina Amin[4], Long Cai[1] ([1]*California Institute of Technology*, [2]*University of California, Los Angeles*, [3]*Stanford University*, [4]*University of California, Riverside*)

1Pos263* (3SEA-2) ラマンイメージングを用いた細胞内の水の可視化とラベルフリー細胞内温度測定への応用
(3SEA-2) Raman imaging of water in a cell and its application to label-free evaluation of intracellular temperature
Toshiki Sugimura, Shinji Kajimoto, Takakazu Nakabayashi (*Grad. Sch. Pham. Sci., Tohoku. Univ*)

1Pos264* Elastin 様ポリペプチドに基づく分子温度センサー
Molecular Thermometer Based on Elastin-Like Polypeptide
Cong Vu, Tetsuichi Wazawa, Takeharu Nagai (*ISIR, Osaka Univ.*)

1Pos265* 新規小分子プローブによるアクチン繊維の可視化と光操作
Visualization and manipulation of actin cytoskeleton by using novel small molecular probes
Takeru Takagi[1], Tasuku Ueno[1], Yusuke Nomura[1], Daisuke Asanuma[2], Yasuteru Urano[1,2,3] ([1]*Grad. Sch. Pharm. Sci., The Univ. Tokyo*, [2]*Grad. Sch. Med., The Univ. Tokyo*, [3]*AMED, CREST*)

1Pos266 (1SCP-6) グルタミン酸受容体を介した植物の長距離 Ca^{2+} シグナル
(1SCP-6) Long-distance Ca^{2+} transmission via glutamate receptor channels in plants
Masatsugu Toyota[1,2] ([1]*Dept Biochem and Mol Biol, Saitama Univ*, [2]*University of Wisconsin-Madison*)

1Pos267 (1SDP-4) Visualization and quantification of biological samples by high-speed atomic force microscope
Hiroki Watanabe[1,2], Koichi Kato[1,2,3], Takayuki Uchihashi[1,4] ([1]*NINS, ExCELLS*, [2]*NINS, IMS*, [3]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [4]*Dept. Phys., Nagoya Univ.*)

1Pos268 (1SGA-8) 細胞内動態をサブセルレベルで制御する温和な NanoHeating 技術
(1SGA-8) A Thermodynamic Tool for Mechanobiology Research: Mild Nanoheating Technology to Alter Subcellular Dynamics
Satoshi Arai[1], Nandus Ferdi[2] ([1]*Res. Inst. Sci. Eng., Waseda Univ.*, [2]*WABIOS*)

1Pos269 クライオ電子顕微鏡により明らかになったノロウイルスの動的構造変化
Dynamic Structural Change of Norovirus Revealed by Cryo-electron Microscopy
Chihong Song[1], Reiko Todaka[2], Masaru Yokoyama[3], Naoyuki Miyazaki[4,5], Kenji Iwasaki[4,5], Kazuhiko Katayama[2], Kazuyoshi Murata[1] ([1]*NIPS*, [2]*Kitasato Univ.*, [3]*NIID*, [4]*IPR, Osaka Univ.*, [5]*Univ. Tsukuba*)

1Pos270 化学発光ビリルビンセンサーの開発
Development of bioluminescent unconjugated bilirubin indicator
Yukino Ito[1], Yoshiyuki Arai[2], Mitsuru Hattori[2], Takeharu Nagai[2] ([1]*Graduate School of Frontier Biosciences, Osaka University*, [2]*The Institute of Scientific and Industrial Research, Osaka University*)

1Pos271 高速原子間力顕微鏡 1 分子計測データを用いた粒子フィルタ法によるリンカー DNA 付きヌクレオソームの動的構造解析
Dynamic structure analysis of nucleosome with linker DNAs by particle filter method using single molecule measurement data by HS-AFM
Sotaro Fuchigami[1,2], Toru Niina[1], Shoji Takada[1,2] ([1]*Grad. Sch. of Science, Kyoto Univ.*, [2]*CREST, JST*)

1Pos272 化学発光トロンビンセンサーの開発
Development of chemiluminescent thrombin sensor toward whole body imaging of living mice
Nae Sugiura[1], Mitsuru Hattori[2], Tomoki Matsuda[2], Takeharu Nagai[2] ([1]*Graduate School of Frontier Biosciences, Osaka univ.*, [2]*Institute of Scientific and Industrial Research, Osaka univ.*)

1Pos273 ESPT 型バイオセンサーの設計
Design of fluorescent biosensors based on Excited State Proton Transfer (ESPT) in the chromophore of a fluorescent protein
Kazunori Sugiura[1,2], Toru Hisabori[2], Shoko Mihara[2], Takeharu Nagai[1] ([1]*ISIR, Univ. Osaka*, [2]*CLS, Tokyo Tech*)

1Pos274 4 次元透過型電子顕微鏡：理論とシミュレーション
4-Dimensional Transmission Electron Microscopy: Theory and Simulation
Kuniaki Nagayama (*N-EM Labos LLC*)

1Pos275 環境の温度変化に対する細胞応答の分子機構
The molecular mechanism of cell response to environmental temperature change
Hiroki Shibata[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ of Tokyo*, [2]*PRESTO, JST*)

1Pos276 相平衡状態にある核小体内領域における核小体構成タンパク質の 1 分子動態と超解像分子局在解析
Single-molecule dynamics and localization of nucleolar proteins in phase-separated compartments of nucleolus
Supanut Sirisukhodom[1], Daiki Matsumoto[1], Yuma Ito[1], Noriko Saitoh[2], Kumiko Sakata-Sogawa[3], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Tech*, [2]*Dept. of Cancer Biol., The Cancer Inst. JFCR.*, [3]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

1Pos277 Development of designable RNA-binding proteins for visualization and manipulation of authentic RNAs in living cells
Akira Takai[1], Yasushi Okada[1,2] ([1]*BDR, RIKEN*, [2]*Univ. of Tokyo, Grad. Sch. of Sci., Dept. of Phys.*)

1Pos278 相分離に関わるヘテロクロマチンタンパク質 HP1α の 1 分子超解像イメージングによる分子局在と動態
Dynamics and localization of Heterochromatin protein 1α involved in phase separation using single-molecule and super-resolution imaging
Takahiro Maeda[1], Yuma Ito[1], Shin-Ya Isobe[2], Chikashi Obuse[2], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Tech*, [2]*Biosci. Grad Sch Sci., Osaka Univ*)

1Pos279 Imaging transcriptional dynamics of the endogenous gene with a bright fluorogenic RNA
Tetsuro Ariyoshi[1], Yasushi Okada[1,2] ([1]*RIKEN BDR, Cell Polarity Regulation*, [2]*Dept. Phys., Grad. Sch. Sci., UTokyo*)

1Pos280 ネクロプトーシスに伴う DAMPs 放出の LCI-S による可視化
Live Cell Imaging for Secretion Activity (LCI-S) of DAMPs Release Accompanying with Necroptosis
Yoshitaka Shirasaki[1,2], Mai Yamagishi[1], Sotaro Uemura[1] ([1]*Dept. of Biological Sciences, Grad School of Science, The Univ. of Tokyo*, [2]*JST PRESTO*)

バイオエンジニアリング／Bioengineering

1Pos281* 単一分子伝導計測に基づく表面上の DNA ハイブリダイゼーションの反応速度論解析
Kinetic investigation of DNA hybridization on surface using single-molecule conductance measurement
Takanori Harashima, Yuki Jono, Tomoaki Nishino (*Sch. Sci., TokyoTech.*)

1Pos282* 制御・情報技術の統合による集団内細胞行動特徴の定量解析
Quantitative analysis of collective cell migration by integration of controlled in vitro experiment and information processing
Asuka Yamaguchi[1], Masakazu Akiyama[2], Ikuhiko Nakase[3], Masaya Hagiwara[4] ([1]*Sch. Sci., Osaka Pref. Univ.*, [2]*MIMS, Meiji Univ.*, [3]*Grad. Sch. Sci., Osaka Pref. Univ.*, [4]*RIKEN, CPR*)

1Pos283* 分散培養心筋細胞と心臓組織片の電気生理学的信号の同期
Synchronization of electrophysiological signal between dispersed cardiomyocytes and cardiac tissue piece
Toru Nakamura, Chiho Nihei, Tomoyuki Kaneko (*Laboratory for Reconstructive Cell biology, Department of Frontier Bioscience, Hosei University, Grad. School of Science and Engineering*)

1Pos284 DNA ナノデバイスを制御する DNA 生成回路の検証
Characterization of DNA Generation Circuits for Controlling DNA Nanodevices
Ken Komiya, Teruya Enomoto, Masayuki Yamamura (*Sch. Comp., Tokyo Tech.*)

1Pos285 Photo-control of Ras nucleotide exchange reaction using the inhibitor peptides modified with spiropyran derivative
Kenichi Taii[1], Nobuyuki Nishibe[1], Kei Sadakane[2], Shinsaku Maruta[1,2] ([1]*Dept. of Bioinfo., Grad. Sch. of Eng., Soka Univ.*, [2]*Dept. of Sus. Inno., Fac. of Sci. and Eng., Soka Univ.*)

結晶成長・結晶化技術／Crystal growth & Crystallization technique

1Pos286 アポフェリチンを使ったマグネタイト単結晶ナノ粒子の作製
Synthesis of single crystal magnetite nanoparticles in apoferritin cavity
Tomoko Kanamaru, Daisuke Katayama, Naoki Takasihima, Takeshi Narusima, **Hideyuki Yoshimura** (*Dpt. Phys., Meiji University*)

その他／Miscellaneous topics

1Pos287 (1SCA-5) 光からエネルギーを合成しタンパク質合成をする人工光合成細胞の構築
(1SCA-5) Artificial photosynthetic cell producing energy for protein synthesis
Samuel Berhanu[2], Takuya Ueda[3], **Yutetsu Kuruma**[1] ([1]*JAMSTEC*, [2]*ELSI, Titech*, [3]*Grad. Sch. of Front. Sci., Univ. of Tokyo*)

1Pos288 人工細胞-生細胞ハイブリッドバイオシステムの創成
Synthesis of artificial/living cell hybrid biosystems
Masamune Morita, Kaoru Katoh, Naohiro Noda (*Biomed. Res. Inst. (BMRI), AIST*)

1Pos289 *Mycolicibacterium smegmatis* のストラクトーム解析
Structome analysis of *Mycolicibacterium smegmatis*
Hiroyuki Yamada[1], Masashi Yamaguchi[2] ([1]*Res. Inst. Tuberculosis., JATA.*, [2]*Mycol. Res. Cent., Chiba*)

1Pos290 Identification of lipid interactions in the transmembrane regions of human Na+, K+-ATPase
Dhani Ram Mahato, Magnus Andersson (*Dept. Che., Ume Univ.*)

2 日目（9 月 25 日（水））／Day 2（Sep. 25 Wed.）
4F 天瑞・ホワイエ／4F TENZUI・Foyer

蛋白質：構造／Protein: Structure

2Pos001* Mycoplasma mobile のモーターを構成するタンパク質 MMOB1620 の構造解析
Structural analysis of MMOB1620 which composes Mycoplasma mobile's motor
Hiroki Sato[1], Aya Kodama[2], Hisashi Kudo[3], Koji Ooka[4], Syunji Suetaka[3], Yuuki Hayashi[3], Munehito Arai[3,4], Makoto Miyata[1,2] ([1]*Graduate School of Science, Osaka City University*, [2]*Faculty of Science, Osaka City University*, [3]*Dept. Life Sci., Univ. Tokyo*, [4]*Dept. Phys., Univ. Tokyo*)

2Pos002* 異なる長さの C 末端領域を持つテロメア繰り返し配列結合タンパク質 AtTRP1 の DNA 結合領域に対する構造研究
Structural studies for DNA binding domain of telomere repeat binding protein, AtTRP-1 with different size of C-terminal region
Shunta Kojima[1], Hayato Morita[2] ([1]*Grad. Sch. Sci., Josai.Univ*, [2]*Fac.Sci., Josai.Univ*)

2Pos003* PaCS-MD によるタンパク質−タンパク質複合体の解離シミュレーション
Protein-protein complexes dissociation simulated by Parallel Cascade Selection Molecular Dynamics
Yoshiki Miyazawa[1], Duy Phuoc Tran[2], Kazuhiro Takemura[2], Akio Kitao[2] ([1]*Grad. Sch. LST.,Tokyo Tech*, [2]*Sch. LST.,Tokyo Tech*)

2Pos004 Deep-Autoencoder に基づいたホモロジーモデリングソフトウェアの開発
Development of Deep-Autoencoder based Homology Modeling software
Masaya Furue, Mitsutaka Nemoto, Lisa Matsukura, Naoyuki Miyashita (*BOST KINDAI Univ.*)

2Pos005 Caged-GTP を用いたがん遺伝子産物 Ras の SACLA, SPring-8, NMR による GTP 加水分解過程の構造変化の解明
Structural changes on GTP hydrolysis of oncogene product Ras revealed by SACLA, SPring-8 and NMR using photo-controllable caged-GTP
Yoshiteru Makino[1], Takashi Kawamura[2], Shigeyuki Matsumoto[1], Eriko Nango[3], So Iwata[3], Takashi Kumasaka[2], Fumi Shima[4] ([1]*Grad. Sch. Med., Kobe. Univ.*, [2]*Protein Cryst. Anal. Div., JASRI.*, [3]*Grad. Sch. Med., Kyoto Univ.*, [4]*Grad. Sch. Sci. Tec. Innov., Kobe Univ.*)

2Pos006 アブラナ科植物の自家不和合性を制御するタンパク質 SRK/SP11 複合体の Rosetta と accelerated MD を用いた構造モデリング
Computational modeling of SRK/SP11 protein complexes using Rosetta and accelerated MD simulations
Yoshitaka Moriwaki[1], Tohru Terada[1,2], Koji Murase[1], Kentaro Shimizu[1] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

2Pos007 粗視化シミュレーションによる CDK4 の構造変化に関する研究
Conformational transition of CDK4 by using coarse-grained simulations
Kazutomo Kawaguchi, Hidemi Nagao (*Inst. Sci. Eng., Kanazawa Univ.*)

2Pos008 Single particle analysis of silkworm lipid transfer protein complex, lipophorin
Shunsuke Kita[1], Kazuhiro Mio[2], Mika Hirose[3], Kenji Iwasaki[4], Naruhiko Adachi[5], Toshio Moriya[5], Masato Kawasaki[5], Katsumi Maenaka[1] ([1]*Fac. of Pharm. Sci., Hokkaido Univ.*, [2]*Operand OIL, AIST*, [3]*IPR, Osaka Univ.*, [4]*TARA, Univ. of Tsukuba*, [5]*SBRC, KEK*)

2Pos009 HIV-2 糖タンパク質の構造機能解析
Structure and functional analysis of human immunodeficiency virus type-2 (HIV-2) envelope glycoprotein
Yuki Anraku[1], Shunsuke Kita[2], Hideo Hukuhara[2], Simon Davis[3], Atsushi Hurukawa[2], Thushan de Silva[3], James Robinson[4], Yuguang Zhao[3], Yvonne Jones[3], David Stuart[3], Juha Huiskonen[3], Sarah Rowland-Jones[3], Katsumi Maenaka[2] ([1]*Grad. Sch. Life Sci., Univ. Hokkaido*, [2]*Faculty of Pharm Sci., Univ. Hokkaido*, [3]*Univ. Oxford*, [4]*Univ. Tulane*)

2Pos010 分子動力学シミュレーションを用いた α シヌクレインアミロイドの構造解析
Structural analysis of α-synuclein amyloids using molecular dynamics simulation
Hiroki Otaki, Yuzuru Taguchi, Noriyuki Nishida (*Grad. Sch. Biomedical Sci., Nagasaki Univ.*)

2Pos011 翻訳後修飾によってシトルリン化したヒト S100A3 蛋白質の擬似体の探索
Exploring the posttranslational modification of human S100A3 protein using citrullination mimics
Kenji Ite[1,2], Kenji Kizawa[3], Kenichi Kitanishi[4], Masaki Unno[1,2] ([1]*Graduate School of Science and Engineering, Ibaraki University*, [2]*Frontier Research Center for Applied Atomic Sciences, Ibaraki University*, [3]*Kao Corporation*, [4]*Department of Chemistry, Faculty of Science, Tokyo University of Science*)

2Pos012 機械学習を用いたタンパク質主鎖構造における 2 面角の再分類
Reclassification of dihedral angles in protein backbone structures using machine learning
Hiroto Murata, George Chikenji (*Dept. Appl. Phys., Nagoya Univ.*)

2Pos013 糖化が LDL の物性に与える影響について
Effect of glycation on the physical properties of low density-lipoprotein
Seiji Takeda[1], Toshihiro Sakurai[1], Shu-Ping Hui[1], Hitoshi Chiba[2] ([1]*Faculty of Health Sciences, Hokkaido Univ.*, [2]*Faculty of Health Science, Sapporo University of Health Sciences*)

2Pos014 酵母プリオン Sup35 の液-液相分離と線維化に対する共溶質の影響
Effects of co-solutes on liquid-liquid phase separation and fibrillization of yeast prion Sup35
Suguru Nishinami[1], Yumiko Ohhashi[2], Kentaro Shiraki[1] ([1]*Pure and Appl. Sci., Univ. Tsukuba*, [2]*Grad. Sch. Sci., Univ. Kobe*)

2Pos015 局所クラスター間構造コンプライアンスに基づくタンパク質の形状に内在する変形伝播特性の解析
Analysis of the Deformation Transmission Properties in Protein Shapes based on the Structural Compliance between Localized Clusters
Keisuke Arikawa (*Fcl. Eng., Kanagawa Inst. of Tech.*)

2Pos016 フェレドキシン構造とそのリバース構造の、PDB における発生頻度が異なるのはなぜか？
Why occurring frequencies of ferredoxin and its reverse fold in PDB are largely different?
Megumi Nakajima, George Chikenji (*Nagoya University Graduate School of Engineering Department of Applied Physics Sasai Laboratory*)

2Pos017 Structural Characterization of β_2 Microglobulin Core Fragments in Amyloid Fibrils using Circular Dichroism Theory and Molecular Dynamics
Koichi Matsuo[1,3], Hirotsugu Hiramatsu[2], Robert W. Woody[3] ([1]*Hiroshima Synchrotron Radiation Center, Hiroshima University*, [2]*Department of Applied Chemistry, National Chiao Tung University*, [3]*Department of Biochemistry and Molecular Biology, Colorado State University*)

2Pos018 B 型肝炎ウイルス(HBV)への逆転写阻害薬剤分子のカプシド内部の自由エネルギー計算
Calculation of free energy of transfer of a reverse transcription inhibitor to the inside of Hepatitis B Virus (HBV) capsid
Ryo Urano[1], Kazushi Fujimoto[1], Yoshimichi Andoh[2], Noriyuki Yoshii[2], Wataru Shinoda[1], Susumu Okazaki[1] ([1]*Grad. Sch. Eng., Nagoya Univ.*, [2]*Center Comput. Sci. Grad. Sch. Sci., Nagoya Univ.*)

2Pos019 アンチパラレル β シート中のジスルフィド結合が分泌タンパク質ホールディングに及ぼす影響
The effect of disulfide bonds in anti-parallel β-sheets on secreted protein folding
Hiromi Suzuki (*School of Agri., Meiji Univ.*)

2Pos020 エンドセリン B 受容体の分子動力学シミュレーション
Molecular dynamics simulations of human endothelin B receptor
Koichi Abe[1], Yoshitaka Moriwaki[1], Kentaro Shimizu[1,2], Tohru Terada[2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

2Pos021 クライオ電子顕微鏡単粒子解析法を用いた KcsA の構造解析
Structural Analysis of KcsA by Cryo-EM Single Particle Analysis
Hiroko Takazaki[1], Hirofumi Shimizu[2], Takuo Yasunaga[1] ([1]*Grad. Sch. Comp. Sci. Syst. Eng., KIT*, [2]*Fac. Med. Sci., Univ. Fukui*)

2Pos022 マスク付きセグメンテーション・フィット法による複数サブユニットの電顕マップへの局所重ね合わせ
Masked segmentation fitting of multiple atomic subunits into a local 3D EM density map
Takeshi Kawabata, Haruki Nakamura, Genji Kurisu (*Institute for Protein Research, Osaka University*)

蛋白質：構造機能相関／Protein: Structure & Function

2Pos023* 糸状仮足の構造変化とアクチンフィラメントの分布の相関
Correlation between structural changes of filopodia and distribution of actin filaments
Miho Nakafukasako, Tomoya Higo, Yusuke V. Morimoto, Takuo Yasunaga (*Grad. Sch. Comp. Sci. Syst. Eng., KIT.*)

2Pos024* Does the secondary site of neuraminidase play any significant role in drug resistance of influenza?
Mohini Yadav[1], Manabu Igarashi[2], Norifumi Yamamoto[1] ([1]*Dept. of Engg., Chiba Inst. Tech.*, [2]*Research Center for Zoonosis Control, Hokkaido Univ.*)

2Pos025* 多次元 NMR 分光法を用いたヤエヤマサソリ由来殺虫性ペプチド毒素 LaIT2 の機能領域の溶液構造解析
Structural studies for the functional domains of insecticidal peptide toxin, LaIT2, with heteronuclear multidimensional NMR spectroscopy
Chiharu Tatsushiro[1], Maiki Tamura[2], Hironori Juichi[3], Masahiro Miyashita[3], Hisashi Miyagawa[3], Shinya Ohki[2], Hayato Morita[1,4] ([1]*Fac Sci., Josai Univ*, [2]*Grad. Sch. Mat Sci., JIAST*, [3]*Grad. Sch. Agr., Kyoto Univ*, [4]*Grad. Sch. Sci., Josai Univ*)

2Pos026* 分子動力学計算で探る p53 の C 末端部位の DNA 結合機構
DNA recognition mechanisms of the p53 C-terminal domain Investigated by MD simulation
Yuta Taira[1], Duy Tran[1], Jacob Swadling[2], Akio Kitao[1] ([1]*Tokyo Tech*, [2]*Univ.Tokyo*)

2Pos027* 天然変性タンパク質 Tau の溶液中における過渡的な凝集原繊維構造形成
Intrinsically disordered protein Tau tends to transiently form a part of the protofilament core structure in the soluble state
Ryosuke Kawasaki[1], Shin-ichi Tate[2] ([1]*Dept. MLS, Grad. Sch. Sci., Hiroshima Univ.*, [2]*Prog. MLS, Grad. Sch. Integr. Sci. for Life, Hiroshima Univ.*)

2Pos028 Single-molecule FRET experiments for investigation of DNA single-strand damage recognition mechanism by PARP-1
Anna Sefer[1], Eleni Kallis[1], Tobias Eilert[1], Mara Guariento[1], Nadine Jakobi[1], David Neuhaus[2], Sebastian Eustermann[3], Jens Michaelis[1] ([1]*Ulm University, Institute of Biophysics, Albert-Einstein Allee 11, 89081 Ulm, Germany*, [2]*MRC Laboratory of Molecular Biology, Francis Crick Avenue, Cambridge CB2 0QH, UK*, [3]*Ludwig-Maximilians-University Munich, Gene Center and Department of Biochemistry, Feodor-Lynen-Strasse 25, 81377 Munich, Germany*)

2Pos029 (2SGP-3) Determination of protonated states for native and mutant structures of HIV-1 protease with indinavir by free energy calculations
Masahiko Taguchi, Ryo Oyama, Masahiro Kaneso, Shigehiko Hayashi (*Kyoto University*)

2Pos030 Crucial role of conformational excitation in enzyme catalysis of Pin1
Toshifumi Mori[1,2], Shinji Saito[1,2] ([1]*IMS*, [2]*SOKENDAI*)

2Pos031 Weighted ensemble simulations of the cis-trans isomerization in Pin1 enzyme using the QM/MM method
Norifumi Yamamoto[1], Kei Moritsugu[2], Yasushige Yonezawa[3], Shin-ichi Tate[4], Hiroshi Fujisaki[5] ([1]*Chiba Tech*, [2]*Yokohama City Univ*, [3]*Kindai Univ*, [4]*Hiroshima Univ*, [5]*Nippon Med Sch*)

2Pos032 Simulation Study on Atomistic and Physicochemical Properties of Amyloid beta 42
Ikuo Kurisaki, Shigenori Tanaka (*System Info., Grad. Schl., Kobe Univ.*)

2Pos033 敵対的生成ネットワーク（GAN）を用いた新規主鎖構造のタンパク質デザイン
Protein design with novel main-chain structure using Generative Adversarial Networks
Takaaki Sato[1], Yoshitaka Moriwaki[1], Tohru Terada[2], Kentaro Shimizu[1,2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

2Pos034 テルペン環化酵素における基質と反応の選択性に関する計算科学的研究
Computational investigation of the substrate and reaction selectivity of terpene cyclases
Masanobu Arita[1], Keiichi Murai[1], Yoshitaka Moriwaki[1], Tohru Terada[2], Tomohisa Kuzuyama[1], Kentaro Shimizu[1,2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

2Pos035 Pin1 由来のタンパク質分解酵素のミクロ化
Micronization of a protease derived from Pin1
Teikichi Ikura, Nobutoshi Ito (*Med. Res. Inst., Tokyo Med. Dent. Univ.*)

2Pos036 緑色硫黄細菌 ferredoxin-NADP$^+$酸化還元酵素と基質間の酸化還元反応の特異性
Unique kinetic behavior in the redox reaction catalyzed by ferredoxin-NADP$^+$ oxidoreductase from green sulfur bacteria
Daisuke Seo (*Grd. Sch. Nat. Sci. Tec., Kanazawa Univ.*)

2Pos037 タンパク質キナーゼへの ATP 競合阻害剤結合の自由エネルギー解析
Free energy analysis of ATP competitive inhibitor-protein kinase bindings
Suyong Re, Hiraku Oshima, Yuji Sugita (*RIKEN Center for Biosystems Dynamics Research*)

2Pos038 フォトンファクトリーにおける生体高分子溶液試料の小角 X 線散乱に関する発展
Progress of Biological Small-Angle X-ray Scattering at the Photon Factory
Kento Yonezawa, Masatsuyo Takahashi, Keiko Yatabe, Yasuko Nagatani, **Nobutaka Shimizu** (*KEK, IMSS, PF*)

2Pos039 抗体修飾ナノニードルを用いた生細胞における中間径フィラメントの可動性解析
Mobility analysis of intermediate filament in a living cell using antibody-functionalized nanoneedle and AFM
Ayana Yamagishi[1,2,5], Moe Susaki[1,2], Mei Mizusawa[1,2], Akira Nagasaki[1], Saku Kijima[3], Q.P. Taro Uyeda[1,4], **Chikashi Nakamura**[1,2,5] ([1]*Biomed. Res. Inst., AIST*, [2]*Dept. Biotechnol. & Life Sci., Grad. Sch. Eng., TUAT*, [3]*Bioproc. Res. Inst., AIST*, [4]*Dept. Phys., Sch. Adv. Sci. Eng., Waseda Univ.*, [5]*PhotoBio-OIL, AIST-Osaka Univ.*)

2Pos040 Investigation on the relationship between cytotoxicity and amorphous oligomers
Punitha Velmurugan, Jannatul Aklima, Yoshihiro Ohta, Yutaka Kuroda (*Tokyo university of Agriculture and Technology*)

2Pos041 In vitro ATPase-based screening of circadian clock mutants of KaiC in cyanobacterial circadian clock system
Dongyan Ouyang[1], Atsushi Mukaiyama[1,2], Yoshihiko Furuike[1,2], Kumiko Miwa[3], Takao Kondo[3], Shuji Akiyama[1,2] ([1]*Inst. Mol. Sci.*, [2]*The Grad. Univ. for Adv. Studies*, [3]*Grad. Sch. of Sci., Nagoya Univ*)

2Pos042 構造蛋白質である HIV-1p17 と p24 の動的と静的構造の解析
Dynamic and rigid structures of HIV-1 p17 and p24 proteins
Chiaki Nishimura (*Fac. Pharm. Sci., Teikyo Heisei Univ.*)

2Pos043 アミロイドベータタンパク質を分解する人工ペプチドの設計
Designing artificial peptides that hydrolyze amyloid beta protein
Yoshihiro Iida, Atsuo Tamura (*Kobe Univ, Grad Sch Sci*)

2Pos044 4 つのイントロン位置は、アスパラギン酸アミノ転移酵素立体構造上で平面を形成する。
Four intron positions form a plane in the tertiary structure of aspartate aminotransferase
Michiko Nosaka (*N.I.T., Sasebo College*)

2Pos045 抗微生物ペプチド Cryptdin-4 の多量体の脂質分子存在下における分子シミュレーションによる観察
Antimicrobial peptide Cryptdin-4 oligomers interacting with lipids observed by molecular dynamics simulations
Takao Yoda (*Nagahama Institute of Bio-Science and Technology*)

2Pos046 ヘム ABC インポーター BhuUV-T の構造変化の自由エネルギー解析
Free energy analysis for the conformational changes of a heme ABC importer BhuUV-T
Koichi Tamura[1], Yuji Sugita[1,2,3] ([1]*RIKEN R-CCS*, [2]*RIKEN TMS*, [3]*RIKEN BDR*)

蛋白質：物性・構造／Protein: Property & Structure

2Pos047* トレハロースによるミオグロビンの構造安定化および酸性条件下でのアミロイド形成からの回復作用
Stabilization of the Myoglobin Structure and Restoration from the Amyloid Formation under Acidic Conditions by Trehalose
Satoshi Ajito[1], Mitsuhiro Hirai[1], Nobutaka Shimizu[2], Noriyuki Igarashi[2] ([1]*Grad. Sch. Sci. Tec., Gunma Univ.*, [2]*KEK*)

2Pos048* 異なる緩衝剤中での抗体の安定性と構造変化の関係
Relation between stability and structure of an antibody in different buffers
Hiroaki Oyama[1], Kanta Enomoto[1], Tetsuo Torisu[1], Susumu Uchiyama[1,2] ([1]*Grad. Sch. Eng, Osaka Univ*, [2]*ExCELLS*)

2Pos049* ショウジョウバエ Argonaute2 の N 末端領域はアミロイド繊維を形成する
N-terminal region of Drosophila Argonaute2 can form amyloid fibrils
Haruka Narita, Makoto F. Kuwabara, Tomotaka Komori, Ryo Murakami, Tomohiro Shima, Mikiko C. Siomi, Soutaro Uemura (*Department of Biological Sciences, Graduate School of Science, The University of Tokyo*)

2Pos050* Dynamics of the helix-coil transition of alanine-based polypeptides detected by nanosecond region fluorescence correlation spectroscopy
Supawich Kamonprasertsuk[1,2], Hiroyuki Oikawa[1,2], Satoshi Takahashi[1,2] ([1]*IMRAM.Tohoku Univ.*, [2]*Grad.Sch.Sci.,Tohoku Univ.*)

2Pos051 MD simulation along with MSM analysis reconstructs LPA_6 binding pathway
Rieko Hirota[1], Ryuichiro Ishitani[1], Mizuki Takemoto[1,2], Osamu Nureki[1] ([1]*Dept. of Biosci., Grad Sch. of Sci., Univ. of Tokyo*, [2]*Present address: Preferred Networks, Inc.*)

2Pos052 加熱による中性 pH での β_2 ミクログロブリンのアミロイド線維形成
Heating-induced amyloid formation of β_2-microglobulin at neutral pH
Masahiro Noji[1], Kenji Sasahara[1], Keiichi Yamaguchi[1], Masatomo So[1], Kazumasa Sakurai[2], Jozsef Kardos[3], Hironobu Naiki[4], Yuji Goto[1] ([1]*IPR, Osaka Univ.*, [2]*IAT, Kindai Univ.*, [3]*Dept. Biochem., ELTE*, [4]*Med. Sci., Univ. Fukui*)

2Pos053 Functional Sensitivity and Mutational Robustness of Proteins
Qianyuan Tang, Tetsuhiro Hatakeyama, Kunihiko Kaneko (*Grad. Sch. Art. & Sci., Univ. Tokyo*)

2Pos054 多次元仮想座標とカップルした分子動力学法を用いた mSin3 複合体の立体構造探索
Conformational sampling of an mSin3 complex using multidimensional virtual-system coupled canonical MD
Tomonori Hayami[1,2], Yoshifumi Fukunishi[3], Yoshifumi Nishimura[4], Junichi Higo[5] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Fron. Biosci., Osaka Univ.*, [3]*molprof, AIST*, [4]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*, [5]*Grad. Sch. Sim. Studies., Univ. Hyogo*)

2Pos055 アデニル酸キナーゼの構造転移の改良カメレオンモデルによる研究
Conformational transition of adenylate kinase studied with the improved chameleon model
Ryota Mori, Masaki Sasai, Tomoki P. Terada (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

2Pos056 Spectroscopic analysis of protein crowded environments using the charge-transfer fluorescence probe ANS
Chikashi Ota[1], **Kazufumi Takano**[2] ([1]*Ritsumeikan Univ.*, [2]*Kyoto Pref. Univ.*)

蛋白質：機能／Protein: Function

2Pos057 (2SHA-6) ダイナミン GTP アーゼはアクチン線維の束化と分散を機械的に制御する
(2SHA-6) Dynamin GTPase mechanically regulates bundling and unbundling of actin filaments
Kohji Takei[1], La The Mon[1], Tadashi Abe[1], Tetsuya Takeda[1], Ikuko Fujiwara[2], Akihiro Narita[3] ([1]*Grad.Sch. Med.Dent. Pharm.Sci., Okayama Univ.*, [2]*Dept. Biol.Facul.Sci., Osaka City Univ.*, [3]*Struct. Biol. Res. Ctr and Divi. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

2Pos058 FRET リアルタイム活性測定系を用いた大腸菌 S2P ファミリー膜内切断プロテアーゼ RseP の kinetics 解析
Kinetic analysis of the proteolytic reaction catalyzed by S2P family intramembrane protease RseP using a FRET-based real-time assay system
Yohei Hizukuri, Yoshinori Akiyama (*Inst. Front. Life Med. Sci., Kyoto Univ.*)

2Pos059 Recognition mechanism of proteins which bind to versatile amino acid sequences
Katsumi Omagari (*Dept. of Virology, Medical School, Nagoya City University*)

2Pos060 呼吸鎖超複合体形成タンパク質によって制御される Cyt *c* のパートナータンパク質認識機構
The mechanism of cytochrome *c*-redox partner proteins recognition regulated by the respiratory supercomplex factor protein
Wataru Sato[1], Koichiro Ishimori[2], Peter Brzezinski[1] ([1]*Stockholm Univ. Fac. of Nat. Sci.*, [2]*Hokkaido Univ. Fac. of Sci.*)

2Pos061 QM/MM 法によるニワトリ卵白リゾチームの糖加水分解反応シミュレーション
QM/MM Study on Hydrolysis of Polysaccharides in Hen Egg-White Lysozyme
Takuya Uto[1], Yoshiki Mitani[2], Toshifumi Yui[2] ([1]*Organization for Promotion of Tenure Track, University of Miyazaki*, [2]*Faculty of Engineering, University of Miyazaki*)

蛋白質：計測・解析／Protein: Measurement & Analysis

2Pos062 幾何学に基づく新しいタンパク質構造解析プログラム
A new program to analyze protein structures based on the geometric context
Anri Terabayashi[1], Momoka Nakamura[1], Kyosuke Sakata[1], Takuya Miyakawa[2], Masaru Tanokura[2], Tohru Terada[3], Masaki Kojima[1] ([1]*Sch. Life Sci., Tokyo Univ. Pharm. Life Sci.*, [2]*Grad. Sch. Agric. Life Sci. Univ. Tokyo*, [3]*III, Univ. Tokyo*)

2Pos063 細胞質中 RAF 分子の構造およびダイマー化状態を捉える 1 分子計測
Conformational and dimeric states of cytosolic RAF detected by single-molecule measurements
Kenji Okamoto[1], Kayo Hibino[2], Yasushi Sako[1] ([1]*RIKEN CPR*, [2]*NIG*)

2Pos064 信号変化の順序決定の自動化およびタンパク質 HD 交換ダイナミクスの振動バンドへの応用
Automatic determination of the sequential order of signal changes and its application to vibrational bands of protein H-D exchange process
Daisuke Miyata[1], Takakazu Nakabayashi[1], Shinichi Morita[2] ([1]*Graduate School of Pharmaceutical Sciences, Tohoku University*, [2]*Graduate School of Science, Tohoku University*)

2Pos065 水素高感度解析を実現するタンパク質中性子回折実験の進歩
Progress of the protein neutron diffractometry to realize hydrogen high sensitivity analysis
Ichiro Tanaka (*Graduate School of Science and Engineering, Ibaraki University*)

2Pos066 Flexible Fitting of Biomolecular Structures to Atomic-Force-Microscopy Images via Biased Molecular Simulations
Toru Niina, Sotaro Fuchigami, Shoji Takada (*Grad. Sch. Sci. Univ. Kyoto*)

2Pos067 Deep convolutional neural networks for identifying cryo-EM grid holes suitable for particle collection
Yuichi Yokoyama[1], Tohru Terada[3], Kentaro Shimizu[2,3], Kazutoshi Tani[4] ([1]*GSII, Univ. Tokyo*, [2]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [3]*III, Univ. Tokyo*, [4]*Grad. Sch. Med., Univ. Mie*)

2Pos068 アクチン線維に張力を発生させると線維の長軸まわりのねじれは減少する
Mechanical stress declined the amplitude of the torsional fluctuations of single actin filaments
Kaoru Okura, Takumi Fukuda, Hitoshi Tatsumi (*Department of Applied Bioscience, Kanazawa Inst. of Technol., Ishikawa, Japan*)

2Pos069 高速 AFM 画像のドリフト除去法の応用
Application of drift elimination method for high-speed AFM images
Shotaro Tsujioka[1], Hideji Murakoshi[2], Mikihiro Shibata[3,4] ([1]*Division of Transdisciplinary Sciences,Graduate School of Frontier Initiative,Kanazawa University*, [2]*National Institute for Physiological*, [3]*WPI Nano Life Science Institute (WPI-NanoLSI),Kanazawa University*, [4]*Institute for Frontier Science Initiative,Kanazawa University*)

蛋白質工学／Protein: Engineering

2Pos070* ポリプロリンロッドの挿入によるドメインスワッピング二量体のデザイン
Poly-proline rod insertion for the design of domain-swapped dimer
Shota Shiga[1], Masaru Yamanaka[2], Wataru Hujiwara[1], Shun Hirota[2], Shuichiro Goda[3], Koki Makabe[1] ([1]*Graduate School of Science and Engineering, Yamagata University*, [2]*Division of Materials Science, Nara Institute of Science and Technology*, [3]*Graduate School of Engineering, Nagasaki University*)

2Pos071* c-Myb-KIX 相互作用を阻害するペプチドの計算機によるデザイン
Computational design of a peptide inhibitor targeting c-Myb-KIX interaction
Shunji Suetaka[1], Yoshiki Oka[1], Tomoko Kunihara[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

2Pos072* 超安定タンパク質構造のゼロからの合理設計とその融合による、GPCR 構造の合理的安定化
Tailor-made design of superstable proteins from scratch for rational stabilization of GPCR
Masaya Mitsumoto[1,2], Nanao Suzuki[3], Ryosuke Nakano[3], Takahiro Kosugi[1,2,4], Takeshi Murata[3], Nobuyasu Koga[1,2,4] ([1]*ExCELLS, NINS*, [2]*SOKENDAI*, [3]*Fac. of Sci., Chiba Univ.*, [4]*IMS, NINS*)

2Pos073* 理論的飽和変異解析によるジヒドロ葉酸還元酵素の高活性化
Improving activity of dihydrofolate reductase by theoretical saturation mutagenesis
Kazuhisa Ohara[1], Yoshiki Oka[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

2Pos074* Engineering of genome editing protein Cas9 that slides along DNA faster and might enable efficient target search
Trishit Banerjee[1,2], Dwiky Rendra Graha Subekti[1,3], Hiroto Takahashi[1], Satoshi Takahashi[1], Kiyoto Kamagata[1] ([1]*IMRAM, Tohoku Uni.*, [2]*Fac. of Sci., Tohoku Uni.*, [3]*Grad. Sch. of Sci., Tohoku Uni.*)

2Pos075 (2SGP-8) 天然変性タンパク質 p53 を標的としたペプチドの人工設計ー液液相分離の制御ー
(2SGP-8) Rational design of peptide targeting intrinsically disordered protein p53—regulation of function and phase-phase separation—
Kiyoto Kamagata[1], Eriko Mano[1], Yuji Itoh[1], Saori Kanbayashi[1], Masaya Honda[1], Ryo Kitahara[2], Tomoshi Kameda[3] ([1]*IMRAM, Tohoku Univ.*, [2]*Coll. Pharmacy Sci., Ritsumeikan Univ.*, [3]*AIRC, AIST*)

2Pos076 膜内・膜外領域の改変によるサーモフィリックロドプシンの熱安定化
Further Thermo-Stabilization of Thermophilic Rhodopsin through Engineering in Intramembrane and Extramembrane Regions
Tomoki Akiyama[1], Naoki Kunishima[2], Masako Hirose[3], Sayaka Nemoto[4], Stoshi Yasuda[4,5,6], Yuki Sudo[7], Takeshi Murata[4,6] ([1]*Grad. Sch. Sci. & Eng., Univ. Chiba*, [2]*RIKEN RSC-Rigaku Collaboration Center*, [3]*Malvern Panalytical division of Spectris Co., Ltd*, [4]*Grad. Sch. Sci., Univ. Chiba*, [5]*Inst. Advanced Energy, Univ. Kyoto*, [6]*Molecular Chirality Research Center, Chiba University*, [7]*Grad. Sch. Med. Dent. Pharm. Sci., Univ. Okayama*)

2Pos077 麹菌菌体外放出系の高機能化
Improvement of exocytotic secretion system of *Aspergillus oryzae*
Mone Kogure, Kensuke Nakajima, Yoshinori Tsuji, Yusuke Matsuda (*Dept.Biosci., Grad.Sch.Sci.Tech., Kwansei Gakuin Univ.*)

2Pos078 細胞特異的ゲノム編集を目的とした核酸をデリバリーする新規一本鎖抗体 scFv の作製
Preparation of a novel single chain variable fragment (scFv) which delivers nucleic acid for cell-specific genome editing
Haruka Nasu[1], Yuji Sato[1], Takashi Tsukamoto[2,3], Takashi Kikukawa[2,3], Makoto Demura[2,3], Tomoyasu Aizawa[2,3] ([1]*Grad. Sch. Life Sci. Hokkaido Univ.*, [2]*Fac. Adv. Life Sci. Hokkaido Univ.*, [3]*GSS, GI-CoRE*)

ヘム蛋白質／Heme proteins

2Pos079 生体反応場におけるシトクロム c 内多核ヘムの空間配置・酸化還元状態の変化
Cellular environment modulates geometry and redox state of deca-heme cofactors in bacterial surface cytochromes
Yoshihide Tokunou[1,2,3], Shingo Hattori[4], Thomas Clarke[5], Liang Shi[6], Kazuyuki Ishii[4], Akihiro Okamoto[2] ([1]*Faculty of Life. Environ. Sci., Uni. Tsukuba*, [2]*NIMS*, [3]*Research Fellow of JSPS*, [4]*Inst. Indust. Sci., Univ. Tokyo*, [5]*Centre for Molecular and Structural Biochemistry, Univ. East Anglia*, [6]*Dept. of Biological Sciences, China Univ. of Geosciences*)

2Pos080 NO-binding and protonation process in the catalytic reaction of the bacterial NO reductase as established by time-resolved spectroscopy
Hanae Takeda[1,2], Tetsunari Kimura[3], Takashi Nomura[1], Takehiko Tosha[2], Yoshitsugu Shiro[1], Minoru Kubo[1] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*RIKEN, SPring-8 Center*, [3]*Grad. Sch. Sci., Kobe Univ*)

2Pos081 Anodized gold surface enables mediator-free bioelectrocatalysis of redox enzymes
Yasuhiro Mie, Yoshiaki Yasutake, Mashiki Ikegami, Tomohiro Tamura (*Bioproduction Res. Inst., AIST*)

膜蛋白質／Membrane proteins

2Pos082* 膜蛋白質複合体の構造ダイナミクスへコレステロールが及ぼす影響に関する分子動力学的解析
Influences of cholesterol on structural dynamics of membrane protein complexes studied by molecular dynamics simulations
Hayato Itaya[1], Kota Kasahara[2], Yoshiaki Yano[3], Katsumi Matsuzaki[3], Takuya Takahashi[2] ([1]*Grad. Sch. Life Sci., Ritsumeikan Univ*, [2]*Coll. Life Sci., Ritsumeikan Univ.*, [3]*Grad. Sch. Pharm.Sci., Kyoto Univ*)

2Pos083* Observation of a β-Hairpin Peptide in α-Hemolysin Nanopore
Misa Yamaji, Ryuji Kawano (*Tokyo University of Agriculture and Technology*)

2Pos084* 表面プラズモン共鳴法を用いた膜タンパク質に特異的な脂質の同定: 脂質の生理機能解明を目指して
Identification of membrane proteins-specific lipids using surface plasmon resonance analysis: For elucidating the physiology of lipids
Masataka Inada[1], Masanao Kinoshita[1], Masayuki Iwamoto[2], Shigetoshi Oiki[2], Nobuaki Matsumori[1] ([1]*Grad. Sch. Sci., Kyushu Univ.*, [2]*Fac. Med. Sci., Univ. Fukui*)

2Pos085 クライオ電子顕微鏡による組換リアノジン受容体の高分解能構造
High-resolution cryo-EM structures of recombinant ryanodine receptors
Takuya Kobayashi[2], Akihisa Tsutsumi[3], Kei Saito[4], Takashi Sakurai[2], Masahide Kikkawa[3], Takashi Murayama[2], **Haruo Ogawa**[1] ([1]*IQB, The Univ. Tokyo*, [2]*Juntendo Univ. Grad. Sch. Med.*, [3]*Grad. Sch. Med., Univ. Tokyo*, [4]*Grad. Sch. Arts Sci., Univ. Tokyo*)

2Pos086 Exploring Structural Dynamics of Bacterial ABC Transporter MsbA by High Speed AFM
Kien X. Ngo, Holger Flechsig, Noriyuki Kodera, Toshio Ando (*WPI Nano Life Science Institute, Kanazawa University*)

2Pos087 Corynebacterial “Force-From-Lipids” mechanosensation for glutamate production
Yoshitaka Nakayama[1], Ken-ichi Hashimoto[2,3], Hisashi Kawasaki[2,3], Boris Martinac[1,4] ([1]*Victor Chang Cardiac Research Institute*, [2]*Biotech. Res. Cen., Univ. Tokyo*, [3]*Collab. Res. Ins. Inno. Microbiol., Univ. Tokyo*, [4]*University of New South Wales*)

2Pos088 The assembly of the trimeric autotransporter transmembrane domain into BamA-embedded nanodiscs
Eriko Aoki, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. of Bioinfo., Soka Univ.*)

核酸結合蛋白質／Nucleic acid binding proteins

2Pos089 CRISPR Cas3 と Cse1 複合体の分子動力学シミュレーション
Molecular dynamics simulations of CRISPR Cas3 and Cse1 complex
Tomohiro Yamaguchi, Yui Taketomo, Naoyuki Miyashita (*BOST KINDAI Univ.*)

2Pos090 Identification of proteins that interact with nucleosomes by Quantitative Proteomics
Lumi Negishi[1], Hiroki Tanaka[2], Rina Hirano[1], Tomoya Kujirai[1], Hitoshi Kurumizaka[1] ([1]*IQB, Univ. Tokyo*, [2]*Grad. Adv. Sci. Eng., Waseda Univ.*)

2Pos091 転写因子の振る舞いとクロマチンのゆらぎの関係性を 1 分子計測によって解析する
Single molecular dynamics of transcription factors are controlled by diffusion movement of chromatin
Kazuko Okamoto[1], Yasushi Okada[1], Kuniya Abe[2], Tomonobu M Watanabe[1] ([1]*RIKEN BDR*, [2]*RIKEN BRC*)

2Pos092 DNA 二重鎖切断に応答した DNA トポイソメラーゼ 2B の核内挙動
Dynamic behavior of DNA topoisomerase 2B in response to DNA double-strand breaks
Ken ichi Yano, Keiko Morotomi-Yano (*IPPS, Kumamoto Univ.*)

核酸／Nucleic acid

2Pos093* 人工細胞デバイス内に封入した長鎖 DNA 1 分子からの遺伝子発現
Gene expression from a single large DNA encapsulated in artificial cell device
Yuto Ochiai[1], Hiroshi Ueno[1], Masayuki Su'etsugu[2], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*Dept. Life Sci., Col. Sci., Rikkyo Univ.*)

2Pos094* 生体ナノポアフィルタを用いた DNA の一分子分離
Separation of a single molecule DNA using biological nanopore filter
Asuka Tada, Ryuji Kawano (*Department of Biotechnology and Life Science, Tokyo University of Agriculture and Technology*)

2Pos095* DETECTION OF DNA-POINT-MUTATION USING BIOLOGICAL NANOPORE
Ping Liu, Keisuke Shimizu, Ryuji Kawano (*Tokyo University of Agriculture and Technology*)

2Pos096 DNA を利用した人工分子モーターの実現を目指して
Towards the realization of artificial molecular motor using DNA molecules
Kohei Arai, Yuki Tsushima, Shoichi Toyabe (*Appl. Phys., Tohoku Univ.*)

2Pos097 染色体脱凝縮シミュレーションによるヒト間期核組織化の理解
Organization of interphase human nucleus via simulated chromosome decondensation
Shin Fujishiro, Masaki Sasai (*Dept. Appl. Phys., Nagoya Univ.*)

2Pos098 クロマチン構造と RNA 輸送経路が協調して形成される過程
Cooperative formation of RNA transporting pathway and chromatin structure
Nozomu Imai, Shin Fujishiro, Masaki Sasai (*Dept. Appl. Phys., Nagoya Univ.*)

2Pos099 染色体のナノスケール 3 次元内部構造を可視化するための液中 3 次元原子間力顕微鏡（3D-AFM）の開発
Development of 3D-AFM to visualize nanometer-scale three-dimensional structures of chromosomes in liquid
Keisuke Miyazawa[1], Makiko Meguro-Horike[2], Shin-ichi Horike[2], Takashi Sumikama[3], Taku Higasayama[1], Masayuki Harada[1], Takeshi Fukuma[1,3] ([1]*Kanazawa Univ.*, [2]*Division of Functional Genomics, Advanced Science Research Center, Kanazawa Univ.*, [3]*Nano Life Science Institute (WPI-NanoLSI), Kanazawa Univ.*)

2Pos100 染色体の 3D-AFM 像と実像の関係の理論的解明
A theoretical study on a relationship between 3D atomic force macroscopy image and structure of chromosomes
Takashi Sumikama[1], Keisuke Miyazawa[4], Adam Foster[1,2,3], Takeshi Fukuma[1,4] ([1]*Nano Life Sicence Institute (WPI-NanoLSI), Kanazawa University*, [2]*Department of Applied Physics, Aalto University*, [3]*Graduate School Materials Science in Mainz*, [4]*Division of Electrical Engineering and Computer Science, Kanazawa University*)

2Pos101 FRET study on sequence-dependent unwrapping of nucleosomal DNA
Tomoko Sunami, Di Luo, Hidetoshi Kono (*MMS, iQLS, QST*)

2Pos102 DNA メチル化に依存したヌクレオソームのスライディング動態の解析
Computational Analysis of the Nucleosome Sliding Dynamics Depending on DNA Methylation
Takeru Kameda[1,2], Miho Suzuki[3], Akinori Awazu[1,4], Yuichi Togashi[1,2,4] ([1]*Department of Mathematical and Life Sciences, Hiroshima University*, [2]*RIKEN Center for Biosystems Dynamics Research.*, [3]*Graduate School of Medicine, Nagoya University*, [4]*Graduate School of Integrated Sciences for Life, Hiroshima University.*)

分子遺伝・遺伝情報制御／Molecular genetics & Gene expression

2Pos103* 微小空間内における 1 分子からの長鎖 DNA 複製
Large DNA amplification from single molecule in micro-sized droplet
Hiroki Sawada[1], Naoki Soga[1], Seia Nara[2], Masayuki Su'etsugu[2], Kazuhito V. Tabata[1], Hiroyuki Noji[1] ([1]*Dept. App. Chem, Univ. Tokyo.*, [2]*Dept. Life. Sci., Sol. Sci., Rikkyo Univ.*)

発生・分化／Development & Differentiation

2Pos104* Piezo1 はリンパ管弁形成過程におけるカルシウムシグナルに必要である
Piezo1 is required for calcium signaling during lymphatic valve morphogenesis
Hiroki Katsuta[1,2], Keiko Nonomura[2], Akemi Kanie[2], Takaki Miyata[1], Toshihiko Fujimori[2] ([1]*Grad. Sch. Med. Nagoya Univ. Cell Biol.*, [2]*NIBB Embryology*)

2Pos105 血管新生における血管内皮細胞の往復運動
Linear reciprocating movement of vascular endothelial cells in angiogenesis
Naoko Takubo[1], Kazuaki Naemura[2], Ryo Yoshida[3], Terumasa Tokunaga[4], Hiroki Kurihara[2] ([1]*Isotope Science Center, Univ. Tokyo*, [2]*Grad. Sch. Med., Univ. Tokyo*, [3]*Inst. Statistical Mathematics*, [4]*Faculty of Computer Science and Systems Engineering, Kyushu Inst. Tec.*)

2Pos106 ドリフトのあるランダム運動がヒトの胚葉形成には必要である
Brownian motion with drift is essential for forming human germ layers
Kenshiro Maruyama, Ryo Kobayashi, Haru Hikita, Tadashi Tsubone, **Kiyoshi Ohnuma** (*Nagaoka University of Technology*)

2Pos107 確率的な細胞たちが協調して正確な大きさの体節を作る仕組み ～分節時計によるノイズキャンセル機構～
Noise-resistant developmental reproducibility in vertebrate somite formation
Naoki Honda[1,2], Ryutaro Akiyama[2], Dini WK Sari[2], Bessho Yasumasa[2], Takaaki Matsui[1,2] ([1]*Grad. Sch. Biostudies., Kyoto Univ.*, [2]*NAIST*)

2Pos108 エネルギー地形アプローチに基づく組織形態形成の多様性と安定性の探求
Exploring variety and robustness in tissue morphogenesis based on energy landscape approach
Yoshitaka Kameo[1,2,3], Hironori Takeda[2], Taiji Adachi[1,2,3] ([1]*IFLMS, Kyoto Univ*, [2]*Grad Sch Eng, Kyoto Univ*, [3]*Grad Sch Bio, Kyoto Univ*)

2Pos109 三次元粒子画像流速測定法によるノード流の解析
Analysis of nodal flow by three-dimensional particle image velocimetry
Atsushi Taniguchi[1,2], Yukinori Nishigami[3], Shigenori Nonaka[1,2] ([1]*Spatiotemp. Reg., NIBB*, [2]*ExCELLS*, [3]*RIES, Hokkaido Univ.*)

筋肉／Muscle

2Pos110 Mechanical stress-responsive membrane remodeling in muscle cells
Kenshiro Fujise, Hiroshi Yamada, Kohji Takei, **Tetsuya Takeda** (*Okayama Univ. Grad. Sch. Med. Dent. Pharm. Sci.*)

2Pos111 無機ポリリン酸存在下でのアクチン重合
Polymerization of actin molecules in the presence of inorganic polyphosphate
Koji Ito, Kuniyuki Hatori (*Grad. Sch. Sci. Eng. Yamagata Univ.*)

2Pos112 Visualization of Ca2+ regulated structural change in muscle thin filament by cryoEM
Yurika Yamada, Keiichi Namba, Takashi Fujii (*Grad. Sch. of Frontier Biosci., Osaka Univ.*)

2Pos113 Fhod3 と cMyBP-C による心筋サルコメアの形成および維持機構
Mechanism of construction and maintenance of cardiac sarcomeres by Fhod3 and cMyBP-C
Wataru Kedouin[1], Riho Takiiwa[1], Nao Shimojo[1], Ryu Takeya[2], Takuo Yasunaga[1] ([1]*Grad. Sch. of Comp. Sci. and Sys. Eng., Kyushu Inst. of Tech.*, [2]*Dept. of Pharma., Univ. of Miyazaki*)

2Pos114* DNA オリガミを用いたナノバネ結合心筋ミオシンフィラメントの 1 分子解析
Single molecule analysis of DNA origami-based cardiac myosin filaments attached with Nanospring
Hiroki Fukunaga[1], Masashi Ohmachi[2], Keisuke Fujita[2], Keigo Ikezaki[3], Toshio Yanagida[1,2], Mitsuhiro Iwaki[1,2] ([1]*FBS, Univ. Osaka*, [2]*BDR, Riken*, [3]*Grad. Sch. Sci., Univ. Tokyo*)

2Pos115* 細胞質ダイニンの運動方向性を左右するアミノ酸の同定
Key residues on cytoplasmic dynein for asymmetric unbinding from microtubule
Shintaroh Kubo[1], Tomohiro Shima[2], Takahide Kon[3], Shoji Takada[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*Grad. Sch. Sci., Tokyo Univ.*, [3]*Grad. Sch. Sci., Univ. Osaka*)

2Pos116* 好熱菌由来 F_1-ATPase の至適生育温度における力学的仕事の測定
The Measurement of Mechanical Work of Thermophilic F_1-ATPase at the Optimum Growth Temperature
Tomoaki Okaniwa[1], Yohei Nakayama[2], Naoya Terahara[1], Eiro Muneyuki[1] ([1]*Dept. Phys., Graduate School of Science and Engineering, Chuo Univ.*, [2]*Dept. Appl. Phys., Graduate School of Engineering, Tohoku University*)

2Pos117* 中間鎖をアンカーとした新しい運動アッセイ法によるクラミドモナス軸糸ダイニン集団の運動特性の計測
Collective motility of Chlamydomonas outer arm dynein measured using its intermediate chain as a scaffold for motility assays
Yuka Matsuda[1], Akane Furuta[2], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2], Ken'ya Furuta[2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst., NICT*)

2Pos118 The combination of high-speed atomic force microscopy and X-ray crystallography reveals rotary catalysis of a shaftless V1 motor
Shintaro Maruyama[1], Motonori Imamura[2], Takayuki Uchihashi[3,4], Kazuya Nakamoto[1], Kenji Mizutani[5], Lica Fabiana Imai[1], Kano Suzuki[1], Yoshiko Ishizuka-Katsura[6], Tomomi Someya-Kimura[6], Mikako Shirouzu[1,6,7], Ichiro Yamato[1,7], Toshio Ando[2], Takeshi Murata[1,8] ([1]*Grad. Sch. Sci., Univ. Chiba*, [2]*WPI Nano Life Sci. Inst., Univ. Kanazawa*, [3]*JST, CREST*, [4]*Dep. Phys., Univ. Nagoya*, [5]*Grad. Sch. Med. Life. Sci., Univ. Yokohama*, [6]*DSSB, RIKEN*, [7]*Ind. Sci. Tokyo Univ. Sci.*, [8]*PREST, JST*)

2Pos119* Measurement of force generation by dynein ensemble on a doublet microtubule obtained from sperm flagella
Takashi Fujiwara[1], Chikako Shingyoji[1], Hideo Higuchi[2] ([1]*Dept. Biol. Sci., Grad. Sch. Sci., Univ. Tokyo*, [2]*Dept. Phys., Grad. Sch. Sci., Univ. Tokyo*)

2Pos120* 1 分子計測による腸内連鎖球菌由来の回転分子モーター V_1-ATPase の化学力学共役機構の解明
Chemo-mechanical coupling scheme of rotary molecular motor *Enterococcus hirae* V_1-ATPase revealed by single-molecule analysis
Tatsuya Iida[1,2], Yoshihiro Minagawa[3], Hiroshi Ueno[3], Fumihiro Kawai[4], Takeshi Murata[5], Ryota Iino[1,2] ([1]*SOKENDAI (The Grad. Univ. for Adv. Stud.)*, [2]*Inst. for Mol. Sci.*, [3]*The Univ. of Tokyo*, [4]*Yamagata Univ.*, [5]*Chiba Univ.*)

2Pos121 (2SDA-7) Dynamic energy landscape of a linear motor chitinase from single-particle tracking trajectories
Kei-ichi Okazaki, Akihiko Nakamura, Ryota Iino (*Institute for Molecular Science*)

2Pos122 アクチンの pH 依存的な荷電状態と分子間相互作用
pH-dependent charge-state and intermolecular interaction of actin
Jun Ohnuki, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

2Pos123 枯草菌べん毛モーターの回転ステップ解析
Step analysis of the *Bacillus* Na^+-driven flagellar motor
Naoya Terahara[1,2], Miku Yoh[3], Eiro Muneyuki[1], Keiichi Namba[2,4], Tohru Minamino[2] ([1]*Dept. Phys., Chuo Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Fac. Human Life Sci., Doshisha Women's Col.*, [4]*BDR/RSC, Riken*)

2Pos124 アクチン繊維の集団運動により形成されるベルトパターンはリング状に変化する
Transformation of belt-like to ring patterns of a quasi-concentrated solution of F-actin driven by myosin-coated surface
Kentaro Ozawa[1], Mikiya Amano[1], Hirotaka Taomori[1], Itsuki Kunita[2], Shigeru Sakurazawa[3], Hajime Honda[1] ([1]*Dept. Bioeng., Nagaoka Univ. Tech.*, [2]*Univ. Ryukyus*, [3]*Future Univ. Hakodate*)

2Pos125 Myosin II decreases the connectivity of an actin network using two different mechanisms depended on concentration of crosslinking protein
Kyohei Matsuda[1], Takuya Kobayashi[1,2], Mitsuhiro Sugawa[1], Masahiko Yamagishi[1], Yoko Y. Toyoshima[1], Junichiro Yajima[1] ([1]*Department of Life Sciences, Graduate School of Arts and Sciences, The University of Tokyo*, [2]*Juntendo University*)

2Pos126 Developing the detection system of the conformational change in rotating flagellar motor by single motor FRET
Takuma Nakagawa, Tatsuya Yamakoshi, Yong-Suk Che, Akihiko Ishijima, **Hajikme Fukuoka** (*Grad. Sch. Frontier Biosci., Osaka Univ.*)

2Pos127 ダイニン・微小管・DNA 折り紙複合体の振動的運動
Oscillatory movement of the dynein-microtubule complex crosslinked with DNA-origami
Shimaa A. Abdellatef[1], Hisashi Tadakuma[2], Yuichi Kondo[3], Kangmin Yan[1], Hideo Higuchi[3], **Keiko Hirose**[1] ([1]*Biomed. Res. Inst., AIST*, [2]*IPR, Osaka Univ.*, [3]*Grad. Sch. Sci., Univ. Tokyo*)

2Pos128 Ca2+濃度変化による真核生物鞭毛軸糸の構造変化の X 線繊維回折解析
Structural changes of Chlamydomonas and Ciona flagellar axonemes coupled with the change in [Ca2+] studied with X-ray fiber diffraction
Kazuhiro Oiwa[1], Hiroyuki Iwamoto[2], Kogiku Shiba[3], Kazuo Inaba[3], Hitoshi Sakakibara[1] ([1]*Adv. ICT Res. Inst., NICT*, [2]*JASRI*, [3]*Shimoda Marine Res. Cent. Univ. Tsukuba*)

2Pos129 新規繊毛ダイニン軽鎖 MOT7 の構造機能解析
Structural/functional analyses on MOT7, a novel light chain of ciliary dynein f/I1
Ryosuke Yamamoto[1], Yuuhei Nakagiri[1], Osamu Kutomi[2], Hiroshi Imai[1], Chihong Song[3], Kazuyoshi Murata[3], Ken-ichi Wakabayashi[4], Takashi Ishikawa[5], Kazuo Inaba[6], Takahide Kon[1] ([1]*Osaka Univ.*, [2]*Univ. of Yamanashi*, [3]*NIPS*, [4]*Tokyo Tech*, [5]*PSI*, [6]*Univ. of Tsukuba*)

2Pos130 全反射赤外分光法を用いた共役イオン結合による Na^+駆動型モーター固定子の構造変化の解明
The cation-induced structural changes in the Na^+-driven flagellar stator studied by ATR-FTIR
Masayo Iwaki[1], Tatsuro Nishikino[2], Hiroyuki Terashima[2], Michio Homma[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Nagoya Univ.*)

2Pos131 2 つのアゾベンゼンを持つフォトクロミック Eg5 阻害剤存在下における Eg5 活性の光制御機構の研究
Study on inhibitory mechanism of kinesin Eg5 with photochromic Eg5 inhibitor composed of two azobenzene
Kei Sadakane[1], Islam MD Alrazi[2], Kenichi Taii[2], Tomisin H. Ogunwa[3], Shinsaku Maruta[1,2] ([1]*Sci. & Eng., Soka Univ.*, [2]*Grad. Sch. Eng., Soka Univ.*, [3]*Grad. Sch. Fisheries and Environmental Sci., Nagasaki Univ.*)

2Pos132 結晶性キチン加水分解酵素は背水の陣で進むブラウニアンモーターである
Crystalline chitin hydrolase is a Burnt-bridge Brownian motor
Akihiko Nakamura[1,2], Kei-ichi Okazaki[1], Tadaomi Furuta[3], Minoru Sakurai[3], Ryota Iino[1,2] ([1]*Institute for Molecular Science*, [2]*SOKENDAI*, [3]*Tokyo Institute of Technology*)

2Pos133 プログラマブルな DNA オリガミによる微小管格子構造の制御
Defining microtubule lattice structure using programmable DNA-origami seeds
Daisuke Inoue, Franky Djutanta, Rizal Hariadi (*Biodesingn Institute, Arizona State Univ.*)

2Pos134 Plus-end directionality is present in the conserved catalytic motor core of kinesin-14 minus-end directed motors
Masahiko Yamagishi, Junichiro Yajima (*Dept. Life Sci., Grad. Arts & Sci., Univ. Tokyo*)

2Pos135 DNA オリガミの分子配置技術を用いたキネシン分子の協調性評価
Evaluating coordination between kinesin motors using DNA origami-based transport complex
Kodai Fukumoto[1], Yuya Miyazono[2], Hisashi Tadakuma[1], Yoshie Harada[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Front. Sci., Univ. Tokyo*)

2Pos136 KIF1A/UNC-104 によるシナプス小胞前駆体輸送の数理モデル
Mathematic modeling of synaptic vesicle precursor transport by KIF1A/UNC-104
Ryo Sasaki[1], Ryota Shinagawa[1], Kazuo Sasaki[1], Shinsuke Niwa[2], Kumiko Hayashi[1,3] ([1]*Dep. Appl. Phys., Grad. Sch. of Eng., Tohoku Univ.*, [2]*FRIS, Tohoku Univ.*, [3]*JST, PRESTO, Tokyo, Japan*)

2Pos137 遺伝性痙性対麻痺を引き起こす変異型ヒト KIF1A の運動特性
Motility characteristics of human KIF1A mutants in hippocampal neurons in relation to hereditary spastic paraplegia
Shiori Matsumoto[1], Kyoko Chiba[2], Shinsuke Niwa[3], Kumiko Hayashi[1,4] ([1]*Dep. Appl. Phys., Grad. Sch. of Eng., Tohoku Univ.*, [2]*Col. Biol. Sci., UC Davis*, [3]*FRIS, Tohoku Univ.*, [4]*PRESTO, JST*)

2Pos138 キネシン 1 の連続的移動距離を決める要因の高速一分子観察
Determinant of the processivity of kinesin-1 as studied using high-speed single-molecule observations
Tsukasa Enomoto[1], Kohei Matsuzaki[2], Michio Tomishige[2] ([1]*Grad. Sch. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*)

細胞生物的課題／Cell biology

2Pos139* 集団運動する神経幹細胞で測定された牽引力の、細胞配向場によるモデリング
Traction Force of Neural Stem Cells under Collective Migration was Modeled using the Orientation Field of Cell Alignment
Masahito Uwamichi[1], Kyogo Kawaguchi[2], Masaki Sano[3,4] ([1]*Dept. of Phys, Univ. of Tokyo*, [2]*RIKEN BDR*, [3]*IIAS*, [4]*TOKYO COLLEGE*)

2Pos140* 細胞集団の 3D 自己組織化: 高分子溶液の水／水ミクロ相分離の活用
Self-Generating 3D Cellular Assembly: Aqueous/Aqueous Micro Droplet as a Non-invasive Scaffold
Ritsuki Ito, Toshifumi Kishimoto, Takahiro Kenmotsu, Koichiro Sadakane, Kenichi Yoshikawa (*Graduate School of Life Medical Science, Doshisha University*)

2Pos141* ナノ粒子を用いた細胞内局所加熱に対する細胞の応答
Responses of cells to local heating in cells using a nanoparticle
Hideaki Ota, Hideo Higuchi (*Phys., Grad. Sci., Univ. Tokyo*)

2Pos142* Quantitation of cell shape by machine learning
Masato Tsutsumi[1], Chikara Furusawa[2,3], Satoshi Sawai[4], Nen Saito[2] ([1]*Grad School of Science, The Univ. of Tokyo*, [2]*Universal Biology Institute, The Univ. of Tokyo*, [3]*Center for Biosystems Dynamics Research, RIKEN*, [4]*Graduate School of Arts and Sciences, The Univ. of Tokyo*)

2Pos143* 細胞接着形態振動を駆動するグラフトポリマー層の垂直/水平変形性とその相関解析
Dual characterizations of vertical/lateral deformation of grafted-polymer layer driving cell-shape oscillation
Sayaka Masaike[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

2Pos144* 鍵アミノ酸残基を含む領域に変異をもつべん毛繊維の in vitro 多型変換実験
In vitro polymorphic transformation of flagellar filaments with mutations in a domain including the key amino acids
Shiori Hirose[1], Hidetoshi Tomaru[1], Yuuka Sashida[1], Yuka Kobayashi[1], Kana Horiguchi[1], Mikako Tsubaki[1], Fumio Hayashi[2], Kenji Oosawa[1] ([1]*Gunma Univ, Grad. Sch. Sci. Technol*, [2]*Gunma Univ, Ctr. Instr. Anal*)

2Pos145* マイコプラズマニューモニエの走流性
Rheotaxis in *Mycoplasma pneumoniae*
Yoshiki Kabata, Daisuke Nakane, Takayuki Nishizaka (*Department of Physics, Gakushuin University*)

2Pos146* 細胞内で LLPS 現象を観察する簡単な方法の開発
Development of a simple method to observe LLPS in cells
Chaieun Kim[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo.*, [2]*PRESTO, JST*)

2Pos147 (2SCP-5) Morphodynamic feature space of migrating cells
Daisuke Imoto[1], Nen Saito[2], **Satoshi Sawai**[1,3] ([1]*Graduate School of Arts and Sciences, University of Tokyo*, [2]*Universal Biology Institute, Graduate School of Science, University of Tokyo*, [3]*Research Center for Complex Systems Biology, University of Tokyo*)

2Pos148 バクテリア集団運動の揺らぎと応答の測定
Measurement of fluctuation and response of bacterial collective motion
Tatsuro Kai, Takahiro Abe, Shuichi Nakamura, Seishi Kudo, Shoichi Toyabe (*Appl. Phys., Tohoku Univ.*)

2Pos149 細胞運動のメカノケミカルモデル
Mechanochemical modelling of crawling cells
Mitsusuke Tarama[1], Kenji Mori[2], Ryoichi Yamamoto[2,3] ([1]*RIKEN BDR*, [2]*Dep Chem Eng, Kyoto Univ*, [3]*IIS, Univ Tokyo*)

2Pos150 Quantitative Analysis of Signal-dependent Cell Cycle Regulation
Kyoichi Ebata, Hiroaki Imoto, Sawa Yamashiro, Mariko Okada (*IPR, Osaka Univ.*)

2Pos151 飢餓状態への過渡期における大腸菌集団の束状凝集と一細胞形態
Bundle structure and single-cell morphology in E. coli populations during transient to a starvation condition
Takuro Shimaya[1], Reiko Okura[2], Yuichi Wakamoto[2], Kazumasa A. Takeuchi[1,3] ([1]*Dept. of Phys., Univ. Tokyo*, [2]*Dept. of Basic Sci., Univ. Tokyo*, [3]*Dept. of Phys., Tokyo Tech*)

2Pos152 Adaptability and robustness of cell migration realized by size-dependent polarity dynamics
Akihiko Nakajima[1,2], Motohiko Ishida[2], Satoshi Sawai[2,3] ([1]*Dept. Gen. Sys. Studies, Grad. Sch. Arts & Sci., Univ. Tokyo*, [2]*Res. Cent. Comp. Sys. Biol., Univ. Tokyo*, [3]*Dept. Basic Sci, Grad. Sch. Arts & Sci., Univ. Tokyo*)

2Pos153 3 次元毛細管構造のトポロジーが血管内皮細胞のシート展開の振る舞いを決定する
Topology of three-dimensional capillary structure determines blood vein sheet extension behavior
Kento Iida[1], Hiromichi Hashimoto[1], Masao Odaka[2], Akihiro Hattori[2], Kenji Yasuda[1,2] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Org. Univ. Res. Initiatives, Waseda Univ.*)

2Pos154 血管内皮細胞の二次元平面構造内における単一細胞の運動特性の観察
Behavior of single vascular endothelial cells in 2D structures
Hiromichi Hashimoto[1], Yuki Yamanaka[1], Ryuji Takano[2], Kento Iida[1], Masao Odaka[3], Kenji Matsuura[3], Akihiro Hattori[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

2Pos155 FRET を用いた「CheYp 濃度変化」と「べん毛モーターの回転方向」の同時計測
Simultaneous measurement of flagellar motor rotation and CheYp concentration via single cell FRET
Tatsuya Yamakoshi, Yong-Suk Che, Akihiko Ishijima, Hajime Fukuoka (*Grad.Sch.Frontier.Osaka Univ.*)

2Pos156 低浸透圧下における金魚ケラトサイト細胞シートの移動速度の上昇
Enhanced movement of fish keratocytes cell-sheet under low osmotic conditions
Naoto Ishijima, Hitoshi Tatsumi (*Human Information Systems Laboratory, Kanazawa Institute of Technology*)

2Pos157 A multi-omic approach to predict gene expression and metabolic functions from label-free spectral imaging of living cells
Arno Germond, Vipin Kumar, Tomonobu M. Watanabe (*RIKEN BDR*)

2Pos158 Conduction Pathway Analysis of Line-Networked Cardiomyocytes By using Multi-Electrode Array System
Tetsuro Yoshida, Tomoyuki Kaneko (*LaRC, FB, Grad. Sci.&Eng., Hosei Univ.*)

2Pos159 原子間力顕微鏡を用いた悪性度が異なるがん細胞の細胞間接着強度の比較
Comparison of intercellular adhesion strengths of cancer cells having different malignancies studied by atomic force microscopy
Kenta Ishibashi[1,2], Tomoko Okada[1], Chikashi Nakamura[1,2,3], Hyonchol Kim[1,2,3] ([1]*Biomed. Res. Inst., AIST*, [2]*Grad. Sch. Eng., Tokyo Univ. Agric. Technol.*, [3]*PhotoBio-OIL, AIST-Osaka Univ.*)

2Pos160 Analysis of Physical Effect on Macrophage with Agarose Microchamber
Tomoyuki Irisawa, Nami Morizino, Tomohiro Saito, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

2Pos161 擬似心臓モデルとしての直線状心筋・線維芽細胞ネットワークの再構成および電気生理学性質の評価
Reconstruction of cardiac tissue-like cell structure and electrophysiological property evaluation
Koki Fujii, Tomoyuki Kaneko (*Grad. Sch. FB LaRC, Hosei Univ.*)

2Pos162 多電極電位システムを用いた赤外線レーザー照射による心臓組織片の拍動変化
Beating rate change of heart tissue piece by infrared laser irradiation using Multi Electrode Array system
Koji Emura, Tomoyuki Kaneko (*LaRC, FB, Grad.Sci&Eng, Hosei Univ.*)

2Pos163 ニワトリ胚由来心筋細胞を低温から温度を上げた際の拍動周期の変化
Beating rate change of chick embryonic cardiomyocytes heated up from low temperature
Kohei Oyama, Wei Wang, Tomoyuki Kaneko (*LaRC, FB, HOSEI unv.*)

2Pos164 心臓組織片の薬剤感受性におけるサイズ依存性の分析
Analysis of sensitivity for drugs depending size of cardiac tissue
Ryohei Kobayashi, Koji Emura, Tomoyuki Kaneko (*LaRC,FB,Hosei Univ.*)

2Pos165 ハイドロゲル上でのマスト細胞の脱顆粒抑制機構の研究
Inhibition of degranulation in mast cells attached to a hydrogel through defective microtubule tracts
Tadahide Furuno, Atsushi Shiki, Satoru Yokawa, Yoshikazu Inoh (*Sch. Pharm., Aichi Gakuin Univ.*)

生体膜・人工膜／Biological & Artificial membrane: Structure & Property

2Pos166 局所麻酔薬によるラフト様/非ラフト様相分離の解消とその機序
Mechanism of the local anesthetics-induced perturbation of raft-like ordered membrane domains
Masanao Kinoshita, Takeshi Chitose, Nobuaki Matsumori (*Kyushu University*)

2Pos167 Comparative study on organizations of human stratum corneum intercellular lipids collected from various body sites
Kenta Moriwaki, Hiromitsu Nakazawa, Satoru Kato (*Grad. Sch. Sci & Tech., Univ. Kwansei Gakuin*)

2Pos168* 回折 X 線ブリンキング法を用いた生細胞上の GPCR 分子内部運動の決定
Determining Intramolecular Motion of GPCRs on Live Cells using Diffracted X-ray Blinking Technique
Masaki Ishihara[1,2], Shoko Fujimiura[2], Kohei Ichiyanagi[3,4], Shunsuke Nozawa[3], Shinichi Adachi[3], Ryo Fukaya[3], Masahiro Kuramochi[1,2], Kazuhiro Mio[2], Yuji Sasaki[1,2] ([1]*Grad Sch. of Fron. Sci., Univ. of Tokyo*, [2]*Univ. of Tokyo - AIST OIL*, [3]*KEK*, [4]*Jichi Med. Univ*)

2Pos169* 大腸菌の封入密度に依存したリポソームの形態変化
Morphological changes of liposomes depending on density of encapsulated E. coli
Mai Hayakawa, Hazuki Terajima, Masahito Hayashi, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

2Pos170* 抗菌ペプチド・ラクトフェリシン B が誘起する細胞膜や脂質膜の急速な膜透過には膜電位が重要な役割を果たす
Membrane potential is vital for rapid permeabilization of plasma membranes and lipid bilayers by the antimicrobial peptide lactoferricin B
Farzana Hossain[1], Md. Mizanur Moghal[1], Md. Zahidul Islam[1], Md. Moniruzzaman[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos171* 細胞透過ペプチド・トランスポータン 10 の単一ベシクル内腔への侵入に対する膜電位の効果とそのメカニズム
Effect of membrane potential on the entry of cell-penetrating peptide transportan10 into the lumen of single vesicles and its mechanism
Md. Mizanur Moghal[1], Md. Zahidul Islam[1], Samiron Kumar Saha[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos172 膜タンパク質の分子内部動態解析技術の開発
Understanding intramolecular dynamics of membrane proteins using X-ray based analysis techniques
Kazuhiro Mio[1], Shoko Fujimura[1], Masaki Ishihara[2], Muneyo Mio[1], Masahiro Kuramochi[2], Hiroshi Sekiguchi[3], Tai Kubo[1], Yuji C. Sasaki[2] ([1]*Operand OIL, AIST*, [2]*Grad. Sch. of Front. Sci., The Univ of Tokyo*, [3]*JASRI*)

2Pos173 リン脂質とアクチンとビーズの互いに結合しないもの同士によるリポソームの形態形成
Liposome morphogenesis by phospholipid, actin filament and polystyrene bead that are not bound to each other
Ryota Kojima[1], Tomo Shibuya[2], Yutaka Sumino[2], Shunsuke Tanaka[1], Masahito Hayashi[1], **Kingo Takiguchi**[1] ([1]*Department of Biological Science, Graduate School of Science, Nagoya University*, [2]*Department of Applied Physics, Tokyo University of Science*)

2Pos174 タンパク質吸着による脂質膜の基板からの剥離
Membrane detachment from substrate induced by protein adhesion
Hiroshi Noguchi (*ISSP, Univ. Tokyo*)

2Pos175 エピガロカテキンガレートが誘起する GUV の破裂のメカニズム
Mechanism of the burst of giant unilamellar vesicles induced by epigallocatechin gallate
Yukihiro Tamba[1], Mika Terada[1], Naoya Sugita[1], Masahito Yamazaki[2] ([1]*Natl Inst Tech, Suzuka Coll*, [2]*Shizuoka Univ*)

2Pos176 平面型人工脂質二分子膜への昆虫細胞由来出芽ウイルスの融合観察
Observation of fusion between baculovirus budded virus envelopes and artificial planar bilayer lipid membrane
Azusa Oshima[1], Nahoko Kasai[1], Hiroshi Nakashima[1], Kanta Tsumoto[2], Koji Sumitomo[3] ([1]*NTT Basic Res. Labs.*, [2]*Mie Univ.*, [3]*Univ. Hyogo*)

2Pos177 赤外レーザー照射によるリポソームの形態変化
Morphological changes of liposomes by infrared laser irradiation
Tomoyuki Kaneko, Akira Oguri, Shunsuke Shiomi, Mai Hayakawa, Masahito Hayashi (*LaRC, FB, Hosei Univ.*)

2Pos178 抗菌ペプチド・マガイニン 2 が誘起するポア形成に対する膜電位の効果
Effect of membrane potential on antimicrobial peptide magainin 2 (mag)-induced pore formation in lipid bilayers
Md. Mamun Or Rashid[1], Md. Mizanur Moghal[1], Moynul Hasan[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos179 蛍光プローブでラベルされていない細胞透過ペプチド・トランスポータン 10 と単一巨大リポソームとの相互作用
Interaction of non-fluorescent probe-labelled cell-penetrating peptide transportan 10 with single giant unilamellar vesicles (GUVs)
Madhabi Shuma[1], Md. Mizanur Moghal[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos180 浸透圧により DOPG/DOPC-GUV に誘起される膜張力の評価
Estimation of Membrane Tension of DOPG/DOPC-GUVs Induced by Osmotic Pressure
Samiron Kumar Saha[1], Sayed Ul Alam Shibly[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

2Pos181 抗菌性オリゴ糖が誘発する脂質二分子膜の多層化
An antibacterial oligosaccharide makes lipid bilayer multi-layered
Ayumi Sumino[1,2], Tatsuya Hagiwara[3], Hatsuo Yamamura[3] ([1]*WPI-NanoLSI, Kanazawa Univ.*, [2]*InFiniti, Kanazawa Univ.*, [3]*Grad. Sch. Eng., Nagoya Inst. Tech.*)

2Pos182 Leaflet-specific lipid diffusions in supported lipid bilayers
Takuhiro Otosu, Shoichi Yamaguchi (*Saitama Univ.*)

2Pos183 インクジェット塗布を用いたパターン化人工生体膜の開発
Inkjet-printed and dried lipid membrane arrays for the biophysical studies and biosensing applications
Yasushi Tanimoto[1], Misato Yamada[2], Fumio Hayashi[3], Kenichi Morigaki[1,2] ([1]*Biosignal, Kobe Univ.*, [2]*Grad. Sch. Agr., Univ. Kobe*, [3]*Grad. Sch. Sci., Univ. Kobe*)

2Pos184 Multiscale molecular dynamics simulations of F-BAR protein Pacsin1: Assembly and curvature preference on lipid membrane
Md. Iqbal Mahmood[1], Hiroshi Noguchi[2], Kei-ichi Okazaki[1] ([1]*Institute for Molecular Science, Okazaki*, [2]*Institute for Solid State Physics, University of Tokyo, Kashiwa, Chiba*)

2Pos185 高速 AFM によるハブ毒液由来のホスホリパーゼ A_2 によって引き起こされる膜分解の動態観察
Membrane degradation dynamics by phospholipase A_2 from snake venom observed by high-speed AFM
Magoto Kamiya[1], Naoko Oda-Ueda[2], Ayumi Sumino[3,4] ([1]*Division of Mathematical and Physical Sciences, Graduate School of Natural Science and Technology, Kanazawa University*, [2]*Department of Pharmaceutical Sciences, Sojo University*, [3]*WPI Nano Life Science Institute (WPI-NanoLSI), Kanazawa University*, [4]*Institute for Frontier Science Initiative, Kanazawa University*)

神経回路・脳の情報処理／Neuronal circuit & Information processing

2Pos186 グラフニューラルネットワークを用いた運動想起時脳波分類
Classification of Motor Imagery Using Graph Neural Networks
Ryo Nakajima[1], Hideo Mukai[2] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci. & Tech., Meiji Univ.*)

2Pos187 運動想起時脳波の時間周波数解析とニューラルネットワークによる識別
Time-Frequency and neural network analysis for classification motor imagery EEG
Azumi Ohno[1], Hideo Mukai[1,2] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci. & Tech., Meiji Univ.*)

2Pos188 深層畳み込みニューラルネットワークを用いたマウスの社会的行動の自動検出
Automated detection of social behavior in mice using deep convolutional neural network
Hideo Mukai[1,2], **Kenji Takemoto**[1] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci. & Tech., Meiji Univ.*)

2Pos189 仮想空間を用いた EEG による運動想起フィードバック訓練システムの構築
EEG-Based motor imagery feedback training system on VR environment
Hideo Mukai[1,2], **Kazuki Kobayashi**[1] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech.,Meiji Univ.*, [2]*Dept. Comp.Sci., Sch. Sci & Tech., Meiji Univ*)

2Pos190 線虫 C.elegans の学習行動を制御する神経回路のシナプス可塑性
Mechanisms of Synaptic Plasticity in a Neural Circuit that Regulates Memory Dependent Behavior in C. elegans
Llian Mabardi, Hirofumi Kunitomo, Hirofumi Sato, Yu Toyoshima, Yuichi Iino (*Tokyo University School fo Science Department of Biology*)

2Pos191 海馬で合成される男性・女性ホルモンやストレスホルモンによる記憶シナプスの早い non-genomic な制御
Rapid non-genomic modulation of synapses by hippocampus-synthesized androgen, estrogen and stress steroid
Suguru Kawato[1,2], Mika Soma[1], Mari Ogiue-Ikeda[1] ([1]*Dep. Cognitive Neuroscience, Fac. Pharma-Science, Teikyo Univ.*, [2]*Dep. Urology, Grad Sch Medicine, Juntendo Univ.*)

行動／Behavior

2Pos192 A leadership-based phase transition in a flocking model with activated and un-activated agents
Sulimon Sattari[1], Tamiki Komatusaki[1], Mikito Toda[2], Sky Nicholson[3], Jason Green[3], Udoy Basak[1] ([1]*Hokkaido University, Research Institute for Electronic Science*, [2]*Nara Women's University*, [3]*University of Massachusetts, Boston*)

光生物学：視覚・光受容／Photobiology: Vision & Photoreception

2Pos193* レチナールを結合するリジンを保存しない微生物型ロドプシンの光応答性機能獲得
Engineering microbial rhodopsin without retinal-binding lysine to gain photosensitive function
Yumeka Yamauchi[1], Masae Konno[1,2], Daichi Yamada[1,3], Kei Yura[4,5,6], Keiichi Inoue[1,7], Oded Béjà[8], Hideki Kandori[1,2] ([1]*Life Sci. Appl. Chem., Nagoya Inst. Tech.*, [2]*OBTRC, Nagoya Inst. Tech.*, [3]*Grad. Sch. Life Sci., Univ. Hyogo*, [4]*Grad. Sch. Hum. Sci., Ochanomizu Univ.*, [5]*Sim. Inf. Bio., Ochanomizu Univ.*, [6]*Sch. Adv. Sci. Eng., Waseda Univ.*, [7]*ISSP, Univ. Tokyo*, [8]*Technion - Israel Inst. Tech.*)

2Pos194* T(6-4)C の同位体標識を用いた(6-4)光回復酵素の低温における DNA 修復中間体の赤外分光測定
Low-temperature FTIR study of the repair intermediates of T(6-4)C/photolyase using isotope labeling
Katsuya Maeda[1], Mai Kumagai[1], Daichi Yamada[2], Yuma Terai[3], Junpei Yamamoto[3], Hideki Kandori[1] ([1]*Nagoya Inst. Tech*, [2]*Univ. Hyogo.*, [3]*Osaka Univ.*)

2Pos195* シネコシスティスハロロドプシン（SyHR）のアニオン輸送における 塩基性アミノ酸の機能的役割
Functional roles of basic amino acids on the anion transport in *Synechocystis* halorhodopsin (SyHR)
Masaki Nakama[1], Keiichi Kojima[1], Marie Kurihara[1], Susumu Yoshizawa[2], Yuki Sudo[1] ([1]*Grad. Sch. of Med. Dent.& Pharm. Sci. Okayama Univ.*, [2]*AORI, UTokyo*)

2Pos196* Molecular characterization of heliorhodopsin from marine giant virus light-dependently infecting to *Emiliania huxleyi*
Ritsu Mizutori[1], Masae Konno[1,2], Keiichi Inoue[1,3], Oded Beja[4], Hideki Kandori[1,2] ([1]*Grad. Sch. Eng., NIT*, [2]*OBTRC, NIT*, [3]*ISSP, Univ. Tokyo*, [4]*Technion-Israel Inst. Tech.*)

2Pos197* 分光学的手法による霊長類青感受性視物質の光反応中間体解析
Photochemical reactions of a primate blue-sensitive pigment by spectroscopic study
Shunpei Hanai[1], Kota Katayama[1], Takuma Sasaki[1], Hiroo Imai[2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Primate Research Institute, Kyoto University*)

2Pos198 (2SFA-6) 微生物型ロドプシンに基づく光遺伝学ツールの探索と開発
(2SFA-6) Exploration and development of microbial rhodopsin-based optogenetic tools
Keiichi Kojima, Yuki Sudo (*Grad. Sch. of Med. Dent. Pharm. Sci., Okayama Univ.*)

2Pos199 ロドプシンクラスター上におけるトランスデューシンの動的過程の高速 AFM 観察
High-speed AFM observation of the dynamic process of transducin on rhodopsin cluster
Kazuhiko Hoshikaya[1], Yasushi Tanimoto[2], Hayato Yamashita[1], Kenichi Morigaki[2,3], Fumio Hayashi[4], Masayuki Abe[1] ([1]*Graduate School of Engineering Science, Osaka University*, [2]*Biosignal research center, Kobe University*, [3]*Graduate School of Agricultural Science, Kobe University*, [4]*Graduate School of Science, Kobe University*)

2Pos200 網膜桿体細胞内円盤膜上での脂質-光受容タンパク質秩序形成の数理モデル
A mathematical model of pattern formation of lipid-photoreceptor proteins on disk membranes of retinal cells
Yukito Kaneshige[1], Yasushi Tanimoto[2], Hiraku Nishimori[1], Kenichi Morigaki[2], Fumio Hayashi[3], Akinori Awazu[1] ([1]*Dept. of Math. & Sci. Hiroshima Univ.*, [2]*Dept. of Agri. Kobe Univ.*, [3]*Dept. of Sci. Kobe Univ.*)

2Pos201 新奇チャネルロドプシン Ts_Rh3 の電気生理学的解析
Electrophysiological analysis of a novel channelrhodopsin Ts_Rh3
Rintaro Tashiro[1], Kumari Sushmita[2], Suneel Kateriya[2], Hideki Kandori[1], Satoshi Tsunoda[1,3] ([1]*Nagoya Institute of Technology*, [2]*Jawaharlal Nehru University*, [3]*JST PRESTO*)

2Pos202 リン酸化ロドプシン·アレスチン複合体は視細胞円板膜切れ込み部に集まる
Phosphorylated-rhodopsin/arrestin complex assembles to disc incisures
Fumio Hayashi[1], Fuko Kueda[2], Kenichi Morigaki[2,3], Keiji Seno[4] ([1]*Kobe Univ, Sci, Biology*, [2]*Kobe Univ, Agri*, [3]*Kobe Univ, Biosignal*, [4]*Hamamatsu Univ Sch Med*)

2Pos203 ボルボックスの光驚動反応における鞭毛運動の照度依存性
Light intensity dependence of adaptive photo-response of Volvox
Yukariko Komasaka, Yoshihiro Murayama (*Department of Applied Physics, Tokyo University of Agriculture and Technology*)

2Pos204 共鳴ラマン分光法と MD + QM/MM 計算を用いたシアノバクテリオクロム発色団の脱プロトン化部位の同定
Identification of the Deprotonated Pyrrole Nitrogen of the Bilin-Based Photoreceptor by Raman Spectroscopy with MD+QM/MM Analysis
Risako Miyoshi[1], Shinsuke Osoegawa[1], Kouhei Watanabe[1], Yuu Hirose[2], Tomotsumi Fujisawa[1], Masahiko Ikeuchi[3], Masashi Unno[1] ([1]*Dept. Chem. & Appl. Chem., Saga Univ.*, [2]*Dept. of Appl. Chem. & Life Sci., Toyohasi Univ. of Tech.*, [3]*Dept. Life Sci. (Biology), Univ. of Tokyo,*)

2Pos205 ビリベルジン結合型シアノバクテリオクロムの遠赤／橙色光変換過程での構造変化の検出
Detection of structural change during far-red/orange reversible photoconversion of biliverdin-binding cyanobacteriaochrome
Yuka Takeda, Keiji Fushimi, Rei Narikawa (*Grad. Sch. Sci., Univ. Shizuoka*)

2Pos206 赤外分光法によって明らかになった色覚視物質とロドプシンの構造ダイナミクスの違い
Different structural dynamics between cone pigments and rhodopsin revealed by FTIR spectroscopy
Takuma Sasaki[1], Kota Katayama[1], Hiroo Imai[2], Hideki Kandori[1] ([1]*Grad. Sch. Eng., Nagoya Inst. Tech.*, [2]*Primate Res. Inst., Kyoto Univ.*)

2Pos207 An Anion Channelrhodopsin with a Naturally Super-Slow Photocycle
Takahiro Kitahara[2], Hina Kurane[3], Chihiro Kikuchi[2], Tomoyasu Aizawa[1,4], Takashi Kikukawa[1,4], Makoto Demura[1,4], **Takashi Tsukamoto**[1,4] ([1]*Fac. Adv. Life Sci., Hokkaido Univ.*, [2]*Grad. Sch. Life Sci., Hokkaido Univ.*, [3]*Sch. Sci., Hokkaido Univ.*, [4]*GSS, GI-CoRE, Hokkaido Univ.*)

2Pos208 赤外分光法により明らかとなった酵素型ロドプシンの構造的特徴
Structural features of enzyme rhodopsins revealed by infrared spectroscopy
Masahito Watari[1], Tatsuya Ikuta[2], Haon Hutamata[2], Daichi Yamada[1], Wataru Shihoya[2], Kazuho Yoshida[1], Yuji Hurutani[1], Satoshi Tsunoda[1,3], Osamu Nureki[2], Hideki Kandori[1] ([1]*Life Sci. Appl. Chem., Nagoya Inst. Tech.*, [2]*Grad. Sch. Sci., Univ. Tokyo.*, [3]*PREST, JST*)

2Pos209 *Mastigocladopsis repens* halorhodopsin の Cl^-ポンプ活性における His166 の重要性
Importance of His166 for Cl^--pump activity of *Mastigocladopsis repens* halorhodopsin
Kento Iwama[1], Yumi Watanabe[1], Takashi Tsukamoto[1,2,3], Tomoyasu Aizawa[1,2,3], Makoto Demura[1,2,3], Takashi Kikukawa[1,2,3] ([1]*Grad. Sch. Life Sci., Hokkaido Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*GSS, GI-CoRE, Hokkaido Univ.*)

2Pos210 時間分解フーリエ変換赤外分光法による KR2 のナトリウムおよびリチウムイオン輸送の分子機構研究
Time-resolved FTIR spectroscopy for studying molecular mechanisms of sodium and lithium ion transportation of *Krokinobacter* rhodopsin 2
Sahoko Tomida[1], Hideki Kandori[1], Yuji Furutani[1,2] ([1]*Nagoya Inst. Tech.*, [2]*Inst. Mol. Sci.*)

2Pos211 Rc-PYP の多量体複合体形成における K72 の役割
Functional role of a residue K72 of Rc-PYP in light dependent oligomeric complex formation process
Yoichi Yamazaki[1], Natsuki Oka[1], Yugo Hayashi[1], Hironari Kamikubo[1,2] ([1]*Div. Mat. Sci. NAIST*, [2]*IMSS,KEK*)

光生物学：光合成／Photobiology: Photosynthesis

2Pos212 FTIR study on the localization of the excited triplet state of chlorophyll in photosystem II
Taichi Hayase[1], Yuichiro Shimada[1], Ryo Nagao[1,2], Takumi Noguchi[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*RIIS, Okayama Univ.*)

2Pos213 緑色イオウ細菌 Chlorobaculum tepidum からの反応中心複合体標品の改良
Improved preparation of the reaction center complex from the green sulfur bacterium *Chlorobaculum tepidum*
Koki Wada[1], Chihiro Azai[2], Tetsuko Nakaniwa[3], Genji Kurisu[3], **Hirozo Oh-oka**[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Coll. Life Sci., Ritsumeikan Univ.*, [3]*Inst. Protein Res., Osaka Univ.*)

2Pos214 Carotenoid glycoside quenches bacteriochlorophyll *a* fluorescence in the photosynthetic reaction center complex of green sulfur bacteria
Chihiro Azai[1], Jiro Harada[2], Takumi Inoue[1], Shogo Fujimoto[3], Shinji Masuda[4], Daisuke Kosumi[3,5] ([1]*Col. Life Sci., Ritsumeikan Univ.*, [2]*Dept. Med. Biochem., Kurume Univ. Sch. Med.*, [3]*Grad. Sch. Sci. & Tech., Kumamoto Univ.*, [4]*Cent. Biol. Res. & Info., Tokyo Inst. Tech.*, [5]*IPPS, Kumamoto Univ.*)

2Pos215 Isolation of the Rieske/cytochrome *b* complex from green sulfur bacteria and interaction of the Rieske protein with cytochrome *c*-556
Hiraku Kishimoto[1], Takahiro Nagaoka[1], Chihiro Azai[2], Risa Mutoh[3], Hideaki Tanaka[4], Yohei Miyanoiri[4], Genji Kurisu[4], Hirozou Oh-oka[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Col. Life Sci. Ritsumeikan Univ.*, [3]*Fac. Sci., Fukuoka Univ.*, [4]*Inst. Protein Res., Osaka Univ.*)

2Pos216 Spectroscopic characterization of a bacteriochlorophyll b-based LH1-RC complexes from thermophilic purple bacterium Blactochloris tepida
Yukihiro Kimura[1], Ryuta Seto[1], Tomoaki Kawakami[2], Rikako Kishi[1], Michie Imanishi[1], Shinichi Takaichi[3], Shinji Takenaka[1], Michael T. Madigan[4], Seiu Otomo[2] ([1]*Grad. Sch. Agri. Sci., Kobe Univ.*, [2]*Ibaraki Univ.*, [3]*Tolyo Univ. of Agri.*, [4]*Southern Illinois Univ.*)

2Pos217 光感受性アデニル酸シクラーゼ OaPAC の活性制御部位の同定
Identification of the activity-regulating site in the photoactivated adenylate cyclase (OaPAC)
Minako Hirano[1], Tomoya Ishido[2], Masumi Takebe[3], Toru Ide[2], Shigeru Matsunaga[3] ([1]*Grad. Sch. Creation Photon Indust.*, [2]*Okayama Univ.*, [3]*Hamamatsu Photonics K.K.*)

2Pos218 Light-dependent structural states of OaPAC
Tomoya Ishido[1], Toru Ide[1], Minako Hirano[2] ([1]*Okayama University*, [2]*GPI*)

2Pos219 タイプ 1 ロドプシンの L/Q スイッチがヘリオロドプシンの波長制御に及ぼす影響
Effects of the L/Q switch on color tuning of heliorhodopsin
Yuta Nakajima, Hideki Kandori (*Grad. Sch. Eng., NIT*)

2Pos220 クリプト藻由来のカチオンチャネルロドプシン Gt_CCR4 のオプトジェネティクスに向けた電気生理学的研究
Study of cation channelrhodopsin Gt_CCR4 from cryptophyte for optogenetics
Shunta Shigemura[1], Shoko Hososhima[1], Hideki Kandori[1], Satoshi Tsunoda[1,2] ([1]*Grad. Sch. Eng., NIT*, [2]*JST PRESTO*)

2Pos221 中赤外レーザーの神経系培養細胞のカルシウム濃度および膜電位への影響
The influence of mid-infrared laser on Ca2+ concentration and membrane potential of neuron-like cells
Yoshiyuki Shimizu, Toyohiko Yamauchi, Tatsuo Dougakiuchi, Gen Takebe (*Hamamatsu Photonics K.K.*)

2Pos222 中赤外光照射による細胞のアポトーシスシグナル誘導
Induction of intracellular apoptotic cell signaling by mid-infrared laser exposure
Gen Takebe, Yoshiyuki Shimizu, Toyohiko Yamauchi, Tatsuo Dougakiuchi (*Hamamatsu Photonics K.K. Central Research Laboratory*)

2Pos223 生体エネルギーを浪費する光駆動内向きプロトンポンプロドプシンの電気生理学研究と光遺伝学への応用
Electrophysiological study and optogenetics application of inward-directed proton-pumping rhodopsin, NsXeR
Satoshi Tsunoda[1,2], Shoko Hososhima[1], Hideki Kandori[1] ([1]*Nagoya Institue of Technology*, [2]*JST PRESTO*)

2Pos224 Photo-regulate small GTPase Ras using photochromic peptide inhibitor
Nobuyuki Nishibe[1], Kenichi Taii[1], Toshio Nagashima[2], Toshio Yamazaki[2], Kazunori Kondoh[1], Shinsaku Maruta[1] ([1]*Depertment of Bioinformatics, Soka University Graduate School of Engineering, Hachioji, Japan*, [2]*Center for Life Science Technologies, RIKEN, Yokohama, Japan*)

放射線生物：活性酸素／Radiobiology & Active oxygen

2Pos225 プリオンペプチド銅錯体のレドックスポテンシャル
Redox potential of copper-binding prion peptide
Shuhei Murakami, Wakako Hiraoka (*Grad.Sch.of Sci.& Tech.,Meiji Univ*)

2Pos226 低酸素下での X 線誘発 DSB の修復効率
Rejoining efficiency of X-ray-induced DSBs in hypoxia
Ryoichi Hirayama, Akiko Uzawa, Yoshiya Furusawa, Sumitaka Hasegawa (*NIRS, QST*)

2Pos227 低線量放射線に曝露された細胞の運命決定における ATM を介した細胞質の放射線応答重要性
Importance of ATM-mediated cytoplasmic radiation response in determining the fate of cells exposed to low-dose radiation
Munetoshi Maeda[1], Hideki Matsumoto[2], Masanori Tomita[3] ([1]*Proton Medic. Res. Div., R&D Dept., WERC*, [2]*Dept. Exp. Radiol. Health Phys., Sch. Med. Sci., Univ. Fukui*, [3]*Radiat. Safety Res. Center, NTRL, CRIEPI*)

2Pos228 酸化ストレスが引き起こす HeLa 細胞ミトコンドリア電子伝達系の機能増幅
Oxidative stress-induced enhancement of mitochondrial electron transport chain in HeLa cells
Wakako Hiraoka, Shuhei Murakami (*Department of Physics, Meiji University*)

生命の起源・進化／Origin of life & Evolution

2Pos229* DNA を自発的に取り込んだ細胞サイズ液滴
Aqueous polymer solutions create stable cell-sized sphere entrapping DNA: A novel scenario of de novo cell
Fumika Fujita[1], Hiroki Sakuta[1], Kanta Tsumoto[2], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Facul. Life Med. Sci., Doshisha Univ.*, [2]*Facul. Eng., Mie Univ.*)

2Pos230* 遺伝子破壊変異の頻度が調節可能な系の構築およびそのゲノム縮小の進化実験への応用
Construction of a Genetic Tool for Tuning Gene-Inactivating Mutations and its Application to Experimental Evolution of Genome Reduction
Yuki Kanai[1], Saburo Tsuru[2], Chikara Furusawa[2,3] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*UBI, Univ. Tokyo*, [3]*BDR, RIKEN*)

2Pos231 Membraneless Polyester Microdroplets as Primordial Compartments at the Origins of Life
Tony Z Jia[1], Kuhan Chandru[1], Yayoi Hongo[1], Rehana Afrin[1], Tomohiro Usui[1], Kunihiro Myojo[2], Po-Hsiang Wang[1], H. James Cleaves[1] ([1]*Earth-Life Science Institute, Tokyo Institute of Technology*, [2]*Tokyo Institute of Technology Department of Earth and Planetary Science*)

2Pos232 情報高分子と連携したベシクルの自己生産
Reproduction of Vesicles coupled with Template Polymerization
Minoru Kurisu[1], Harutaka Aoki[1], Takehiro Jimbo[1], Yuka Sakuma[1], Masayuki Imai[1], Sandra Luginbuhl[2], Peter Walde[2] ([1]*Grad. Sch. Sci., Univ. Tohoku*, [2]*dep. Material, ETH*)

2Pos233 Laboratory evolution of Escherichia coli reveals constrained evolutionary states for antibiotic resistance
Junichiro Iwasawa[1], Tomoya Maeda[2], Takaaki Horinouchi[2], Chikara Furusawa[1,2,3] ([1]*Dept. of Physics, Univ. of Tokyo*, [2]*RIKEN BDR*, [3]*UBI, Univ. of Tokyo*)

2Pos234 Fitness landscape of antibiotic-resistance evolution
Masayoshi Hiranaka[1], Nen Saito[2], Chikara Furusawa[2,3] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*UBI, Univ. Tokyo*, [3]*BDR, RIKEN*)

2Pos235 Relationship between fluctuation of single-enzyme activity and evolvability
Hiroshi Ueno[1], Morito Sakuma[1], Yoshihiro Minagawa[1], Kazuhito Tabata[1], Kentaro Miyazaki[2,3], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*AIST*, [3]*Grad. Sch. Front. Sci., Univ. Tokyo*)

ゲノム生物学／Genome biology

2Pos236 数理モデルとライブイメージングデータを用いた分裂酵母間期核内構造の解析
Analysis of fission yeast interphase intranuclear structure by mathematical model and live imaging data
Yuki Takayama[1], Hisamichi Senda[2], Koki Ito[3], Hiraku Nishimori[3], Masaru Ueno[3], Akinori Awazu[3] ([1]*Grad. Sch. Sci., Univ. Hiroshima*, [2]*Grad. Sch. Advanced Sciences of Matter, Univ. Hiroshima*, [3]*Grad. Sch. Integrated Sciences for Life, Univ. Hiroshima*)

2Pos237 Dynamical chromatin organization during transcription
Ashwin S. Selvarajan[1], Kayo Hibino[2], Yuji Itoh[2], Kazuhiro Maeshima[1,2], Masaki Sasai[2] ([1]*Dept of Applied Physics, Nagoya University, Nagoya, Japan*, [2]*Structural Biology Center, National Institute of Genetics, Mishima, Shizuoka, Japan*)

生命情報科学／Bioinformatics

2Pos238 機械学習を用いたタンパク質部分配列の構造及び機能の解析
Analysis of structural and functional propensities for subsequences of proteins by using machine learning
Ryohei Kondo[1], Kota Kasahara[2], Takuya Takahashi[2] ([1]*Grad. Sch. Life Sci., Ritsumeikan Univ.*, [2]*Coll. Life Sci., Ritsumeikan Univ.*)

2Pos239 GPCR ー G タンパク質の結合選択性に関わる部位同定および結合 G タンパク種予測への応用
Determination of key regions relating GPCR-Gprotein coupling selectivity and their application for predicting coupling G-protein kinds
Mayu Kawamura[1], Risako Kasado[1], Tomomi Manaka[1], Ryuuji Shinozaki[1,2], Masami Ikeda[3], **Makiko Suwa**[1,2] ([1]*Aoyamagakuin Univ. College of Sci. and Eng.*, [2]*Aoyamagakuin Univ. Grad. School. Sci. and Eng.*, [3]*AIST AIRC*)

2Pos240 デノボデザインによる新規 αβ 型蛋白質フォールドの探査
Exploration of novel alpha-beta protein folds by de novo design
Shintaro Minami[1], Rie Koga[1], George Chikenji[2], Toshihiko Sugiki[3], Naohiro Kobayashi[4], Nobuyasu Koga[1] ([1]*NINS, ExCELLS*, [2]*Grad. Shc. of Eng., Nagoya Univ.*, [3]*Inst. for Prot. Res., Osaka Univ.*, [4]*RIKEN, RSC*)

2Pos241 アンサンブルドッキングによるタンパク質の相互作用面の解析
Analysis of protein interaction surfaces using ensemble rigid-body docking process
Nobuyuki Uchikoga[1], Yuri Matsuzaki[2] ([1]*Dept. of Network Design, Sch. of Interdiscip. Math. Sci., Meiji Univ.*, [2]*ToTAL, TITech*)

2Pos242 転写因子 Med26 における天然変性蛋白質認識メカニズムの分子動力学的検討
Molecular dynamics study for elucidation of recognition mechanism of intrinsically disordered proteins by transcription factor Med26
Satoshi Goto[1], Takuya Takahashi[2], Kouta Kasahara[3] ([1]*Coll Life Sci., Ritsumeikan Univ*, [2]*Prof. Univ.Ritsumei*, [3]*Ass.Prof. Univ.Ritsumei*)

数理生物学／Mathematical biology

2Pos243* (2SCP-6) 上皮メカノケミカル動態の同定
(2SCP-6) System identification of mechano-chemical epithelial sheet dynamics
Yoshifumi Asakura[1], Yohei Kondo[2], Kazuhiro Aoki[2], Naoki Honda[1] ([1]*Grad. Sch. Biostudies, Univ. Kyoto*, [2]*Div. Quantitative Biol. ExCELLS, NIBB.*)

2Pos244* 細胞周期の不均一性に関する網羅的数理モデルの構築
A comprehensive model of heterogeneous cell cycle responses
Hiroaki Imoto, Kyouichi Ebata, Shigeyuki Magi, Suxiang Zhang, Mariko Okada (*IPR, Osaka Univ.*)

2Pos245* 真性粘菌変形体の走磁性とその探索行動への寄与
Magnetotaxis of Physarum plasmodium and its contribution to exploration
Michinori Muro, Hiroshi Sato, Tomohiro Shirakawa (*Natl Def Acad Japan*)

2Pos246 (2SHP-5) 細胞内のインスリン様成長因子-I（IGF-I）シグナルは振動する
(2SHP-5) Cellular insulin-like growth factor-I (IGF-I) signal can be oscillated
Masato Masuda, Fumihiko Hakuno, Shin-Ichiro Takahashi (*Dep. App. Ani. Sci., Grad. Sch. Agr. Lif. Sci., The Univ. Tokyo*)

2Pos247 BOID における第 4 のルール：他個体への注目は一定時間毎に一定の確率で失われる
The fourth rule of BOID: attention to the other individuals is lost with fixed probability every fixed time
Tomohiro Shirakawa, Hiroshi Sato, Takuya Matsuo (*Natl. Def. Acad. Japan*)

2Pos248 An Information-theoretic approach toward identifying the leader(s) and aggregation place in Dictyostelium Discoideum colony
Udoy Sankar Basak[1], Sulimon Sattari[1], Kazuki Horikawa[2], Tamiki Komatsuzaki[1] ([1]*Hokkaido University*, [2]*Tokushima University*)

2Pos249 細胞内共生進化への理論的アプローチ
Theoretical approach to evolution of intracellular symbiosis
Sakura Aoki, Kunihiko Kaneko (*The University of Tokyo Graduate School of Arts and Scienses*)

2Pos250 Asymptotic expansion of a stochastic FitzHugh-Nagumo model
Takanobu Yamanobe (*Sch. Med., Hokkaido Univ.*)

非平衡・発生リズム／Nonequilibrium state & Biological rhythm

2Pos251 光応答性を持った C. elegans の集団運動
Collective motion of optically susceptible C. elegans
Ken Nagai[1], Hiroshi Ito[2], Takuma Sugi[3] ([1]*JAIST*, [2]*Kyushu Univ.*, [3]*Shiga Univ. Med. Sci.*)

2Pos252 逆ミセルを用いたフレーリッヒ凝縮の研究 (I)
Studies of Frohlich condensation using reverse micelles. I
Hiroshi Murakami (*QST*)

2Pos253 孤立した粒子の運動解析を基とした、回転する自走粒子の集団運動を表す数理モデルの推定
Estimation of mathematical model representing collective motion of rotating self-propelled particle
Tadashi Sakaguchi[1], Kazuhiro Oiwa[2], Hitoshi Sakakibara[2], Ken Nagai[1] ([1]*JAIST*, [2]*NICT*)

2Pos254 抗原識別における確率的ノイズの役割
A role of stochastic noise in ligand discrimination
Masashi K. Kajita, Kazuyuki Aihara, Tetsuya J. Kobayashi (*IIS, University of Tokyo*)

2Pos255 Theoretical model of dynamics of epithelial tissue with cellular chirality
Takaki Yamamoto[1], Tetsuya Hiraiwa[2], Tatsuo Shibata[1] ([1]*RIKEN BDR*, [2]*Mechanobiology Institute, National University of Singapore*)

計測／Measurements

2Pos256* 細胞内温度場は高分子に依存する
Intracellular temperature field depends on polymers
Masaharu Takarada[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ of Tokyo*, [2]*PRESTO, JST*)

2Pos257* インフルエンザウイルス多様性解析に向けた、多次元デジタル計測技術の開発
Multi-Dimensional (MD) digital assay for analysis of influenza virus heterogeneity
Shingo Honda[1], Yoshihiro Minagawa[2], Kazuhito V. Tabata[2], Hiroyuki Noji[2] ([1]*Dept. Bioeng., Grad. Sch. Eng., Univ. Tokyo*, [2]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*)

2Pos258* Development of a method for quantitative profiling of microRNAs in single exosomes
Cinya Chung, Ryo Iizuka, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

2Pos259* 光ファイバーを用いた蛍光相関分光法の開発とエクソソーム研究への応用
Development of optical fiber based fluorescence correlation spectroscopy and application to exosome study
Misato Osaka[1], Johtaro Yamamoto[2,3], Masataka Kinjo[3] ([1]*Grad. Sch. of Life Sci., Hokkaido Univ.*, [2]*Biomed. Res. Inst., AIST.*, [3]*Fac. of Adv. Life Sci., Hokkaido Univ.*)

2Pos260* 細胞内温度変動に関与する分子のスクリーニング法の開発
Development of a method to screen molecules related to intracellular temperature variation
Takashi Mitsubori[1], Kohki Okabe[1,2], Fumi Kano[3], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*PRESTO, JST*, [3]*IIR, Tokyo Inst. of Tech.*)

2Pos261 Optical tweezers with red laser for new applications
Tomohiro Masuda, Wataru Nakashima, Kazuki Nakajima, Shin Yamaguchi, Takashi Sagawa, **Yuichi Inoue** (*SIGMAKOKI CO., LTD.*)

2Pos262 赤外超解像顕微鏡によるガチョウおよびペンギン羽毛の内部構造観察
Orientation-sensitive molecular imaging of keratin proteins of goose and penguin feathers by an IR super-resolution micro-spectroscopy
Hirona Takahashi, Koki Kimura, **Makoto Sakai** (*Faculty of Science, Okayama University of Science*)

2Pos263 ミクロゲルの粘弾性特性の測定
Viscoelastic measurement of a biopolymer microgel by microcapillary aspiration
Atsushi Sakai[1,2], Yoshihiro Murayama[1], Miho Yanagisawa[2] ([1]*Tokyo Univ. of Agri. & Technol.*, [2]*Komaba Inst. Sci., The Univ. Tokyo.*)

バイオイメージング／Bioimaging

2Pos264* 高速イオン伝導顕微鏡による表面電荷のマッピング
Mapping of surface charge by high speed ion conductance microscopy
Shusei Kaihatsu[1], Kazuki Shigyo[2], Toshio Ando[2], Shinji Watanabe[2] ([1]*Grad. Sch. Math. & Phys., Kanazawa Univ.*, [2]*WPI-NanoLSI, Kanazawa Univ.*)

2Pos265* マニピュレーター付き高速 AFM スキャナーの改良
Improvement of high-speed AFM scanner with manipulator
Jun Takano[1], Shun Aoki[2], Kazuki Shigyo[3], Shinji Watanabe[3], Toshio Ando[3,4], Noriyuki Kodera[3,4] ([1]*Grad. Sch. Math. & Phys., Kanazawa Univ.*, [2]*Sch. Math. & Phys., Kanazawa Univ.*, [3]*WPI-NanoLSI, Kanazawa Univ.*, [4]*CREST, JST*)

2Pos266* タンパク質オリゴマー分布イメージング：生細胞内で空間的に不均一なオリゴマーの分布可視化に向けて
Protein Oligomer Imaging: towards Visualization of Spatially Heterogeneous Oligomer Distribution in Living Cell
Ryosuke Fukushima[1], Johtaro Yamamoto[2,3], Masataka Kinjo[2] ([1]*Grad. Sch. of Life Sci., Hokkaido Univ.*, [2]*Fac. of Adv. Life Sci., Hokkaido Univ.*, [3]*Biomed. Res. Inst., AIST*)

2Pos267* A ratiometric bioluminescent indicator for water hardness in living specimen
Md Nadim Hossain[1,2], Ryuichi Ishida[2], Mitsuru Hattori[1,2], Tomoki Matsuda[1,2], Takeharu Nagai[1,2] ([1]*Graduate School of Engineering, Osaka University*, [2]*ISIR, Osaka University*)

2Pos268 (2SHP-4) 大気圧走査電子顕微鏡 ASEM による骨組織再構築の水中免疫電顕法と cryo-TEM 観察
(2SHP-4) Observation of unstained bone tissues and immuno-EM in liquid by ASEM and cryo-TEM
Chikara Sato[1], Shinya Sugimoto[2], Yuri Hatano[1], Mari Sato[1], Eiko Sakai[3] ([1]*Biomedical Res. Inst., AIST*, [2]*Dept. Bacteriol., The Jikei Univ. Sch. Med.*, [3]*Dental Pharmacology, Nagasaki Univ.*)

2Pos269 (2SEA-5) G1 期酵母細胞核内における核酸分布の XFELX 線回折イメージング
(2SEA-5) Distribution of nucleic acids in yeast nucleus of G1 phase visualized by X-ray diffraction imaging using X-ray free electron laser
Masayoshi Nakasako[1,2], Takahiro Yamamoto[1,2], Amane Kobayashi[1,2], Mao Oide[1,2], Koji Okajima[1,2], Yuki Takayama[1,2,3], Tomotaka Oroguchi[1,2], Masaki Yamamoto[2] ([1]*Keio University*, [2]*RIKEN*, [3]*University of Hyogo Prefecture*)

2Pos270 ホウレンソウグラナ膜における光化学系 II 分子間相互作用の高速 AFM による解析
HS-AFM imaging and analyses of intermolecular interaction of photosystem II in grana membrane from spinach
Daisuke Yamamoto (*Fuc. Sci. Fukuoka Univ.*)

2Pos271 光照射でタンパク質機能阻害・細胞死を誘導する単量体光増感緑色蛍光タンパク質の開発
Monomeric green fluorescent protein based photosensitizer for photo-inducible protein inactivation and cell death
Tomoki Matsuda[1], Yemima Dani Riani[1], Kiwamu Takemoto[2], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*Grad Sch. of Med., Yokohama City Univ.*)

2Pos272 RNA ポリメラーゼ II により制御されるクロマチンダイナミクス
Chromatin dynamics regulated by RNA polymerase II
Yuji Itoh, Michael Babokhov, Kayo Hibino, Kazuhiro Maeshima (*NIG*)

2Pos273 電子顕微鏡トモグラフィ像への構造フィッティング
Fitting atomic structure to Electron Microscopy Tomography
Yuki Mori, Suguru Kato, Toru Niina, Shoji Takada (*Kyoto University*)

2Pos274 高輝度なポジティブ型光スイッチング蛍光タンパク質 Kohinoor 2.0 の開発
Development of a highly-bright positively reversibly photoswitchable fluorescent protein Kohinoor 2.0 for super-resolution microscopy
Tetsuichi Wazawa[1], Shusaku Uto[1], Kazunori Sugiura[1], Shunsuke Maeda[2], Katsumasa Fujita[2], Takashi Washio[1], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ*, [2]*Grad Sch of Engin, Osaka Univ*)

2Pos275 プラズモニックナノ粒子を用いた高速マルチカラー生体 1 分子イメージング
Multi-color and high-speed imaging of single biomolecules with plasmonic nanoparticles
Jun Ando[1,2], Akihiko Nakamura[1,2], Mayuko Yamamoto[1], Ryota Iino[1,2] ([1]*IMS, NINS*, [2]*SOKENDAI*)

2Pos276 Characterizing the spatio-temporal heterogeneity on biomolecular concentration, mobility and local environment in live cells
Sho Oasa[1], Aleksandar J. Krmpot[1,2], Stanko N. Nikolic[1,2], Lars Terenius[1], Rudolf Rigler[1,3], Vladana Vukojevic[1] ([1]*Dept. of Clin. Neurosci., Center for Mol. Med., Karolinska Inst.*, [2]*Inst. of Physics, Univ. of Belgrade*, [3]*Dept. of Med. Biochem. and Biophys., Karolinska Inst.*)

2Pos277 Mapping of mechanical property on live cell surface by scanning ion conductance microscope
Satoko Kitazawa[1], Linhao Sun[2], Ayako Housaka[2], Takahiro Watanabe-Nakayama[2], Hiroki Konno[2], Mikihiro Shibata[2], Shinji Watanabe[2] ([1]*Grad. Sch. Math. &Phys., Kanazawa Univ.*, [2]*WPI-NanoLSI, Kanazawa Univ.*)

2Pos278 定量的 ATP イメージングを用いた細胞の代謝状態の空間的相関の解析
Spatial correlation of metabolic states in mammalian cells revealed by quantitative single-cell ATP imaging
Hideyuki Yaginuma[1,2], Yasushi Okada[1,3] ([1]*BDR, Riken*, [2]*Grad. Sch. of Eng., Univ. of Tokyo*, [3]*Grad. Sch. of Sci., Univ. of Tokyo*)

2Pos279 局所特徴モーフィングした補間による深さ解像度の低い 3 次元画像の改善
Feature based local morphing improved interpolation of 3D stack images at low depth resolution
Yutaka Ueno[1], Takashi Kawasaki[2], Totai Mitsuyama[1] ([1]*AIST Tokyo*, [2]*AIST Kansai*)

2Pos280 スマートフォン発光顕微鏡による 1 細胞イメージング
Smartphone based chemiluminescence microscope for single cell imaging
Mitsuru Hattori[1], Sumito Shirane[2], Kuniaki Nagayama[2], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*Life is small. Company*)

2Pos281 鱗翅目昆虫の変態過程における極微弱バイオフォトン発光の連続画像計測
Continuous imaging of biophoton emission of lepidopterous insects during
Shoko Usui[1], Mika Tada[2], Masaki Kobayashi[1] ([1]*Grad. Sch. Elec., Tohtech*, [2]*Center for General Education., Tohtech*)

2Pos282 Construction of a millisecond structured illumination microscope and its application to ultrafast super-resolution live cell imaging
Shinji Kajimoto[1], Tomu Suzuki[1], Narufumi Kitamura[2], Mayumi Takano[2], Naoko Furusawa[3], Yasushi Nakano[3], Kohsuke Gonda[2], Takakazu Nakabayashi[1] ([1]*Grad. Sch. Pharm. Sci., Tohoku Univ.*, [2]*Grad. Sch. Med., Tohoku Univ.*, [3]*Konica Minolta Inc.*)

バイオエンジニアリング／Bioengineering

2Pos283* DNA を用いた反応拡散系のプログラムによるハイドロゲル中におけるパターン形成
Programmable reaction-diffusion system using synthetic DNA for pattern formation in hydrogel medium
Keita Abe, Ibuki Kawamata, Shin-ichiro Nomura M., Satoshi Murata (*1Department of Robotics, Graduate School of Engineering, Tohoku University, Japan*)

2Pos284* マイクロドロップレットを用いた G タンパク質共役型受容体ペプチドアゴニスト探索法のフィジビリティスタディ
Feasibility study of the method to obtain peptide agonists for G protein-coupled receptors using water-in-oil microdroplets
Anna Matsueda[1], Takashi Sakurai[1], Ryo Iizuka[1], Yasuyuki Nakamura[2,3], Jun Ishi[2,3], Akihiro Kondo[2,3], Dong Hyun Yoon[5], Tetsushi Sekiguchi[5], Syuichi Syoji[4], Soichiro Tsuda[6], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Eng. Biol. Res. Cent., Kobe Univ.*, [3]*Grad. Sch. of Sci., Technol. and Innov., Kobe Univ.*, [4]*Dept. of Nanosci. and Nanoeng., Waseda Univ.*, [5]*Res. Org. for Nano & Life Innov., Waseda Univ.*, [6]*On-chip Biotechnol. Co., Ltd.*)

2Pos285* 高分子分解酵素産生微生物の取得のための液滴の変形能を利用したスクリーニング法
Deformability-based microfluidic droplet screening to obtain microbes producing macromolecule-degrading enzymes
Mikihisa Muta[1], Kai Saito[1], Ryo Iizuka[1], Wataru Kawakubo[2], Dong Hyun Yoon[3], Tetsushi Sekiguchi[3], Shuichi Shoji[3], Mei Ito[4], Yuji Hatada[4], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Dept. of Nanosci. and Nanoeng., Waseda Univ.*, [3]*Res. Org. for Nano & Life Innov.*, [4]*Dept. of Life Sci. and Green Chem., Saitama Inst. of Technol.*)

2Pos286 カップ形状微小電極の開発と 1 細胞発現分子計測への応用
Development of cup-shaped microelectrode and its application for detection of expressed biomolecules in single cell level
Airi Kuriyama[1,2], Tomoyuki Kamata[1], Dai Kato[1], Naoshi Kojima[1], Shohei Yamamura[3], **Hyonchol Kim**[1,2,4] ([1]*Biomed. Res. Inst., AIST*, [2]*Grad. Sch. Eng., Tokyo Univ. Agric. Technol.*, [3]*Health Res. Inst., AIST*, [4]*PhotoBio-OIL, AIST-Osaka Univ.*)

2Pos287 太陽光に依存しない細胞内生物発光による光合成
Sunlight independent plant cell photosynthesis by self-contained bioluminescence
Kenji Osabe[1], Megumi Iwano[2], Ryuichi Nishihama[2], Kazushi Suzuki[1], Sakiko Ishida[2], Tomomi Kaku[1], Takayuki Kohchi[2], Takeharu Nagai[1] ([1]*Osaka Univ., I.S.I.R.*, [2]*Kyoto Univ., Grad. Sch. Biost.*)

2Pos288 (2SEP-7) グラフェン電界効果トランジスタとフェムトリットルチャンバーを用いたデバイ遮蔽を超える電気的バイオセンシング
(2SEP-7) Electrical Biosensing beyond the Debye Screening Length Using Graphene Field-Effect Transistor in Femtoliter Microchamber
Takao Ono[1], Yasushi Kanai[1], Koichi Inoue[1], Yohei Watanabe[2], Shin-ichi Nakakita[3], Toshio Kawahara[4], Yasuo Suzuki[4], Kazuhiko Matsumoto[1] ([1]*ISIR, Osaka Univ.*, [2]*Kyoto Pref. Univ of Med.*, [3]*Kagawa Univ.*, [4]*Chubu Univ.*)

3 日目（9 月 26 日（木））／Day 3（Sep. 26 Thu.）
4F 天瑞・ホワイエ／4F TENZUI・Foyer

蛋白質：構造／Protein: Structure

3Pos001 Dynamic docking between a flexible enzyme and its inhibitor using multicanonical MD simulations and binding free energy calculations
Narutoshi Kamiya[1], Gert-Jan Bekker[2] ([1]*Sim. Stu., Univ. Hyogo*, [2]*IPR, Osaka Univ.*)

3Pos002 Biological Structure Model Archive: 計算機で得られた生体分子モデルのアーカイブ
Biological Structure Model Archive: An archive for computationally obtained data
Gert-Jan Bekker, Takeshi Kawabata, Genji Kurisu (*Osaka University, IPR*)

3Pos003 Nanodisc に再構成した好熱菌由来 V-ATPase の単粒子解析
Single-particle analysis of the lipid nanodisc-reconstituted V-type ATPase/synthase from *Thermus thermophilus*
Atsuko Nakanishi[1], Jun-ichi Kishikawa[1], Kaoru Mitsuoka[2], Ken Yokoyama[1] ([1]*Faculty of Life Sci. Kyoto Sangyo Univ.*, [2]*Res. Ctr. for UHVEM. Osaka Univ.*)

3Pos004 Prediction of ligand distribution around a protein by 3D-RISM theory
Masataka Hamano, Masatake Sugita, Takeshi Kikuchi, Fumio Hirata (*Dept. Bioinfo., Coll. Biosci., Ritsumeikan Univ*)

3Pos005 アミノ酸配列情報からのフラボヘモグロビンのフォールディング機構予測
Prediction of the folding mechanism of flavohemoglobin based on average distance statitistical method
Maho Osugi, Takeshi Kikuchi (*Dept. Bioinfo., Coll. Biosci., Ritsumeikan Univ.*)

3Pos006 分布推定アルゴリズムによる単粒子解析投影パラメーター決定
Determination of projection parameters in single particle analysis using Estimation of Distribution Algorithms
Nobuya Mamizu[1,2], Takuo Yasunaga[1] ([1]*Kyushu Institute of Technology*, [2]*SYSTEM IN FRONTIER INC.*)

3Pos007 連続滴定小角 X 線散乱測定を用いた KaiC に対する KaiA の滴定挙動解析
Titration analysis of KaiA for KaiC using continuous titration small angle X-ray scattering
Risako Aoyama[1], Yoichi Yamazaki[1], Kento Yonezawa[2], Atsushi Mukaiyama[3], Yugo Hayashi[1], Sachiko Toma-Fukai[1], Nobutaka Shimizu[2], Shuji Akiyama[3], Hironari Kamikubo[1,2] ([1]*Div. Mat. Sci., NAIST*, [2]*IMSS, KEK*, [3]*CIMoS, IMS*)

3Pos008 Folding properties prediction of ribonuclease and chymotrypsin based on inter-residue average distance statistics
K M Ahsanul Kabir, Takeshi Kikuchi (*Dept. Bioinfo., Grad. Sch. of Life Sci., Ritsumeinkan Univ., Computational Biomolecular Chemistry lab.*)

3Pos009 The off-axis rotor of *Enterococcus hirae* V-type ATPase by Volta phase contrast cryo-EM
Raymond N. Burton-Smith[1], Jun Tsunoda[1], Yu Yamamori[2], Naoyuki Miyazaki[3], Fabiana L. Imai[4], Chihong Song[1], Kentaro Tomii[2], Kenji Iwasaki[3], Junichi Takagi[5], Hiroshi Ueno[7], Takeshi Murata[4], Ryota Iino[6], Kazuyoshi Murata[1] ([1]*NIPS*, [2]*AIST*, [3]*Univ. Tsukuba*, [4]*Chiba Univ.*, [5]*Osaka Univ.*, [6]*IMS*, [7]*Univ. Tokyo*)

3Pos010 GTP 結合型および GDP 結合型微小管におけるチューブリン C 末端構造分布差の分子動力学計算による解析
Simulation study for conformational difference of tubulin C-terminal tails in GTP-bound and in GDP-bound microtubule
Takuma Todoroki[1], Yukinobu Mizuhara[2], Jun Ohnuki[2], Mitsunori Takano[2], Koji Umezawa[1,3] ([1]*Grad. Sch. Of Sci. & Tech., Shinshu Univ.*, [2]*Grad. Sch. of Adv. Sci. & Eng., Waseda Univ.*, [3]*IBS, Shinshu Univ.*)

3Pos011 距離依存誘電率および溶媒接触表面積を取り込んだ SAAP 力場を用いたトリプケージの分子シミュレーション
Molecular simulation of Trp-cage using the SAAP force field with Distance-Dependent Dielectric and Solvent Accessible Surface Area
Koji Yoshida, Taku Shimosato, **Michio Iwaoka** (*Tokai Univ., Depart. Chem.*)

3Pos012 asymmetry of psi-loop motifs
Koki Fukuda, George Chikenji (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

3Pos013 溶液 NMR による水素結合の直接観測
Direct observation of hydrogen bonds by solution NMR
Taiki Koizumi, Hiroki Nakajima, Yutaka Ito, Masaki Mishima (*Grad. Sch. Sci., TMU*)

3Pos014 (3SHA-5) STAP-2 により Breast tumor kinase が活性化する機構の解明
(3SHA-5) Molecular basis of Breast tumor kinase by an adaptor protein, STAP-2
Junki Nakasako[1], Yuki Matsuo[2], Ryo Kanda[2], Yoshino Tanaka[2], Min Yao[3], Tadashi Matsuda[2], Katsumi Maenaka[2], Toyoyuki Ose[2,3,4] ([1]*Graduate school of Life Science*, [2]*Faculty of Pharm.*, [3]*Faculty of Advanced Life Science, Hokkaido University*, [4]*JST PRESTO*)

3Pos015 分子シミュレーションで探る angiotensin II type 1 receptor（AT1R）の活性化メカニズム
Activation mechanism of the angiotensin II type 1 receptor (AT1R) explored by molecular simulations
Yuichiro Kanamori, **Tadaomi Furuta**, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech*)

3Pos016 クライオ電子顕微鏡による多剤排出ポンプ複合体 MexAB–OprM の構造解析
The wild-type structures of MexAB-OprM multidrug efflux pump revealed by cryo-electron microscopy
Kenta Tsutsumi[1], Ryo Yonehara[1], Etsuko Ishizaka-Ikeda[1], Naoyuki Miyazaki[1,2], Shintaro Maeda[1,3], Kenji Iwasaki[1,2], Atsushi Nakagawa[1], Eiki Yamashita[1] ([1]*IPR, Univ. Osaka*, [2]*TARA, Univ. Tsukuba*, [3]*The Scripps Research Inst.*)

3Pos017 IgG 抗体の Fv フラグメントはドメイン運動に伴う水和構造変化によって抗原認識ループの構造変化を制御する
Domain motion of Fv-fragment in antibody immunoglobulin G controls conformation of antigen-recognizing loop through hydration structure
Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2] ([1]*Faclt. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*)

3Pos018 3D–RISM 理論を応用した溶液中における Met–enkephalin の構造揺らぎの解析
Analysis of structural fluctuations of Met-enkephalin in the solution phase by means of 3D-RISM theory
Masatake Sugita[1], Fumio Hirata[2] ([1]*Dept. of Bioinfo., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Toyota Phys. & Chem. Res. Inst.*)

3Pos019 網羅的構造解析による酵素機能関連天然変性蛋白質の探索
Exploration of disordered proteins associated with enzymatic functions by comprehensive structural search
Satoshi Omori[1], Hafumi Nishi[1], Kengo Kinoshita[1,2] ([1]*GSIS, Tohoku Univ.*, [2]*ToMMo, Tohoku Univ.*)

3Pos020 A local structural environment descriptor toward evaluating impact of rare variants in humans on protein structures and functions
Atsushi Hijikata, Masafumi Shionyu, Tsuyoshi Shirai (*Nagahama Inst. Bio-Sci. Tech.*)

3Pos021 A new MD integration enabling large time step from accurate temperature and pressure evaluations
Jaewoon Jung[1,2], Hiraku Oshima[3], Kento Kasahara[3], Chigusa Kobayashi[1], Takaharu Mori[2], Yuji Sugita[1,2,3] ([1]*RIKEN Center for Computational Science*, [2]*RIKEN Cluster for Pioneering Research*, [3]*RIKEN Center for Biosystems Dynamics Research*)

3Pos022 Protein interactions in the *in vitro* cyanobacterial circadian clock system revealed by SDSL-ESR
Risa Mutoh[1], Takahiro Iida[1], Hiroyuki Mino[2], Masahiro Ishiura[3] ([1]*Faculty of Sci, Fukuoka Univ.*, [2]*Dep. Sch. of Sci., Nagoya Univ.*, [3]*CGR, Nagoya Univ.*)

3Pos023 Investigation of formation mechanism of Prx high molecular weight complexes
Mami Jindai[1], Rino Sasaki[1], Noriyuki Kodera[2], Toshio Ando[2], **Hiroki Konno**[2] ([1]*Sch. of Nat. Syst., Coll. of Sci. & Eng., Kanazawa Univ.*, [2]*WPI Nano Life Sci. Inst. (WPI-NanoLSI), Kanazawa Univ.*)

3Pos024 Time-localised Predictions of Conformational Transitions in Protein Dynamics
Ryuhei Harada[1], Vladimir Sladek[2], Yasuteru Shigeta[1] ([1]*CCS, Unv. of Tsukuba*, [2]*Slovak Academy of Sciences*)

3Pos025 フレキシブルドッキングによる結合自由エネルギーと速度定数計算
Calculation of binding free energy and kinetic rates with flexible protein docking
Duy Tran, Akio Kitao (*Tokodai, Grad. Life Sci. Tech.*)

3Pos026 Evolutionary diversity of Kai-protein clock system in cyanobacteria
Atsushi Mukaiyama[1,2], Dongyan Ouyang[1], Yoshihiko Furuike[1,2], **Shuji Akiyama**[1,2] ([1]*IMS*, [2]*SOKENDAI*)

3Pos027 Hierarchical classification method of protein-protein interfaces based on their secondary structures
Takashi Fujii, Kazuo Fujiwara, Masamichi Ikeguchi (*Grad. Sch. of Eng., Soka Univ*)

3Pos028 タンパク質ダイナミクスに対する拡散マップ法の適用
Applications of a diffusion map method to protein dynamics
Hiroto Kikuchi[1], Ayori Mitsutake[2], Hiroshi Fujisaki[1] ([1]*Dept. of Phys. Nippon Med. Sch.*, [2]*Dept. of Phys. Meiji Univ.*)

3Pos029 myPresto, computer-aided drug development software
Shinji Iida[1,11], Ikuo Fukuda[2], Junichi Higo[2], Kota Kasahara[3], Takashi Kurosawa[4], Tadaaki Mashimo[5], Kiyotaka Misoo[5], Yoshinori Wakabayasi[6], Ryuta Murakami[1,7], Chisato Kanai[1,7], Yusuke Sugihara[8], Mitsuhito Wada[1], Hironori Nakamura[9], Yoshifumi Fukunishi[10] ([1]*NNPC*, [2]*Grad. Sch. Sim. Hyogo Univ.*, [3]*Col. of Life Sci., Ritsumeikan Univ.*, [4]*Hitachi Solutions East Japan, Ltd.*, [5]*IMSBIO Co., Ltd.*, [6]*BY-HEX LLP*, [7]*INTAGE Healthcare Inc. Drug Discovery Suport Department*, [8]*Fujitsu Kusyu Systems Ltd.*, [9]*Biomodeling Research Co., Ltd.*, [10]*AIST, molprof*, [11]*JBiC*)

3Pos030 十二量体フェリチン様 Dps の希土類バイオミラリゼーションにおける構造基盤
Structural basis of rear-earth metal biomineralization in dodecameric ferritin-like protein, Dps
Mitsuhiro Okuda[1,2], Pretre Gabriela[1], Kornelius Zeth[3] ([1]*CIC nanoGUNE*, [2]*Ikerbasque*, [3]*Roskilde University*)

3Pos031 演題取り消し

蛋白質：物性・構造／Protein: Property & Structure

3Pos032 天然構造が極めて類似したタンパク質の熱変性：遷移構造が異なる分子起源の理論研究
A theoretical study for thermal unfolding of proteins with quite similar native structure and different transition structures
Ken Tomihara, Takashi Yoshidome (*Department of Applied Physics, Tohoku University*)

3Pos033 分子動力学法と機械学習を用いたテトラペプチドの凝集性の評価
Evaluating aggregation propensity of tetra-peptide using MD and machine learning
Yoichi Kurumida, Yutaka Saito, Tomoshi Kameda (*AIRC, AIST*)

3Pos034 Molecular evolution of the structure elements in the TIM barrel family proteins
Yasumichi Takase[1], Yugo Hayashi[1], Yoichi Yamazaki[1], Sachiko Toma-Fukai[1], Hironari Kamikubo[1,2] ([1]*Div. Mat. Sci., NAIST*, [2]*IMSS KEK*)

3Pos035 メタノール中アラメチシンのヘリックス構造熱安定性
Thermal stability of helical conformation of alamethicin in methanol
Yoshinori Miura (*Center for Advanced Instrumental Analysis, Univ. Kyushu*)

3Pos036 ヨウ素染色を用いた種の異なるインスリンアミロイド構造の識別及びシード依存的構造伝播の追跡
Structural difference and its seed-dependent propagation of human/bovine insulin amyloid fibrils as detected by iodine staining
Keisuke Yuzu, Eri Chatani (*Grad. Sch. Sci., Kobe Univ.*)

3Pos037 Examination of the possibility of the formation of transmissible transthyretin amyloid fibrils by the use of proteolysis
Misato Matsumura[1], Naoki Yamamoto[2], Keiichi Yamaguchi[3], Masatomo So[3], Yuji Goto[3], Eri Chatani[1] ([1]*Grad. Sch. Sci., Kobe Univ.*, [2]*Fac. Med., Jichi Med. Univ.*, [3]*Inst. Protein Res., Osaka Univ.*)

3Pos038 The mechanisms underlying the inhibition of amyloid formation by polyphenol
Yuto Kimura, Masatomo So, Yuji Goto (*IPR, Osaka Univ.*)

蛋白質：機能／Protein: Function

3Pos039 In vitro assembly of metabolon by liquid-liquid phase separation
Tomoto Ura, Kentaro Shiraki (*Pure and Appl.Sci.,Univ.Tsukuba*)

3Pos040 Binding properties of heart-type Fatty-Acid-Binding Protein proved by 1,8-ANS displacement assay
Shun Tokudome[1], Mai Nomura[1], Fumio Hayashi[2], Shigeru Sugiyama[3], Shigeru Matsuoka[4], Michio Murata[5], Masashi Sonoyama[1,6,7] ([1]*Grad Sch. Sci-Tech., Univ. Gunma*, [2]*Ctr. Inst. Analysis, Univ. Gunma*, [3]*Sch. Sci-Tech., Univ. Kochi*, [4]*Grad Sch. Med., Univ. Ooita*, [5]*Grad Sch. Sci., Univ. Osaka*, [6]*GIAR., Univ. Gunma*, [7]*GUCFW., Univ. Gunma*)

3Pos041 混雑環境系におけるタンパク質ーリガンド結合機構の速度論的解析
Crowder effects on a protein-ligand binding process
Kento Kasahara[1], Suyong Re[1], Hiraku Oshima[1], Isseki Yu[4], Grzegorz Nawrocki[5], Michael Feig[5], Yuji Sugita[1,2,3] ([1]*RIKEN BDR*, [2]*RIKEN R-CCS*, [3]*RIKEN CPR*, [4]*Maebashi Inst. of Tech.*, [5]*Michigan State Univ.*)

蛋白質：計測・解析／Protein: Measurement & Analysis

3Pos042 マニフォールドラーニングを用いた低温電子顕微鏡 4 次元イメージング法の確立に向けて
Toward constructing a four-dimensional imaging technique for cryo-electron microscopy with manifold learning
Takashi Yoshidome (*Department Applied Physics, Tohoku University*)

3Pos043 投影方向に偏りがある低温電子顕微鏡像の全投影方向の像の回復に向けて
Toward a recovery of images from all irradiation directions using cryo-electron microscopy data with biased irradiation directions
Ryota Kojima, Takashi Yoshidome (*Department of Applied Physics, Tohoku University*)

3Pos044 バイオ医薬品のプロセス開発および QC における MAM のためのワークフロー駆動型プラットフォームソリューション
A workflow driven platform solution for MAM-based critical quality attribute monitoring of biotherapeutics in process development and QC
Kenji Hirose[1], Maki Terasaki[1], Shota Nakamura[1], Nilini Ranbaduge[2], Henry Shion[2], Ying Qing Yu[2], Min Du[2], Weibin Chen[2] ([1]*Nihon Waters*, [2]*Waters*)

3Pos045 カチオンイオン交換と超小型飛行時間型質量分析計を用いたオンライン IEX-MS による モノクローナル抗体チャージバリアントの特性解析とモニタリング
Online IEX-MS Characterization and Monitoring of mAb Charge Heterogeneity Us ing an Optimized Cation Exchange Resin and Compact TOF MS
Maki Terasaki[1], Shota Nakamura[1], Kenji Hirose[1], Samantha Ippoliti[2], Wang Qi[2], Yu Ying Qing[2], Lauber Matthew A.[2] ([1]*Nihon Waters K. K.*, [2]*Waters Corporation*)

3Pos046 DFA イオンペア試薬、高密度フェニル結合充填剤による ADC の高感度 LC-MS プロファイリング
High sensitivity LC-MS profiling of ADC with difluoroacetic acid ion pairing and a high coverage phenyl-bonded stationary phase
Hiroko Iwasaki[1], Kenji Hirose[1], Jennifer Nguyen[2,3], Jacquelynn Smith[4], Olga V. Friese[4], Jason C. Rouse[4], Daniel P. Walsh[2], Matthew A. Lauber[2] ([1]*Nihon Waters K. K.*, [2]*Waters Corporation*, [3]*Univ. of Copenhagen*, [4]*Biotherapeutics Pharm, Sci., Pfizer WRD*)

3Pos047 ポリリン酸による鶏リゾチームの凝集
Polyphosphate-induced aggregation of hen lysozyme
Kenji Sasahara, Keiichi Yamaguchi, Masatomo So, Yuji Goto (*IPR Osaka university*)

3Pos048 超小型高分解能質量分析計の導入によるバイオ医薬品開発の課題の解決
Meeting Challenges of Implementing Accurate-Mass Mass Spectrometry for Biotherapeutic Development in Regulated/non-Regulated Environments
Shota Nakamura[1], Taiji Kawase[1], Maki Terasaki[1], Kenji Hirose[1], Henry Shion[2], Mellisa Ly[2], Nilini Ranbaduge[2], Ximo Zhang[2], Yun Alelyunas[2], Jonathan Pugh[2], Robert Lewis[2], Jill Lord[2], Mark Halifax[2], Nick Tomczyk[2], Dale Cooper-Shepherd[2], Laetitia Denbigh[2], Ying Qing Yu[2], Jason Rouse[3], Weibin Chen[2] ([1]*Nihon Waters K.K.*, [2]*Waters Inc.*, [3]*Pfizer Inc.*)

蛋白質工学／Protein: Engineering

3Pos049 珪藻殻への有用タンパク質提示発現による機能性材料開発
Protein display on the silica frustules of a marine diatom
Natsuki Onishi, Kensuke Nakajima, Yoshinori Tsuji, Yusuke Matsuda (*Dept.Biosci., Grad.Sch.Sci.Tech., Kwansei Gakuin Univ.,*)

3Pos050 Development of the engineered trimeric single-chain Fv fragment of the therapeutic antibody
Takashi Tadokoro, Kota Nakamura, Harumi Tsuboi, Katsumi Maenaka (*Faculty of Pharmaceutical Sciences, Hokkaido University*)

3Pos051 多様なユビキチン鎖のロバストな合成法
Robust synthesis methods of various ubiquitin chains
Takumi Suzuki, Takahiro Aizu, Yutaka Ito, Masaki Mishima (*Grad. Sch. Sci., TMU*)

3Pos052 De novo design of protein structures with Ploop-motif for ATP binding
Hiroko Yamada[1], Kengo Nakamura[1], Takahiro Kosugi[1,2,3], Nobuyasu Koga[1,2,3] ([1]*SOKENDAI*, [2]*NINS IMS*, [3]*NINS ExCELLS*)

ヘム蛋白質／Heme proteins

3Pos053 金属タンパク質における酸化還元電位の第一原理計算法の開発
An ab initio method of evaluating redox potential for metalloprotein
Cheng Cheng, Shigehiko Hayashi (*Kyoto Univ*)

3Pos054 ウシミトコンドリア由来酸素還元酵素の活性型の単量体構造
Monomeric structure of an active form of respiratory oxygen reductase from bovine mitochondria
Kyoko Shinzawa-Itoh[1], Takashi Sugimura[2], Tomonori Misaki[2], Yoshiki Tadehara[1], Shogo Yamamoto[1], Makoto Hanada[1], Naomine Yano[1], Tetsuya Nakagawa[3], Shigefumi Uene[1], Takara Yamada[3], Hiroshi Aoyama[4], Eiki Yamashita[5], Tomitake Tsukihara[1,5], Shinya Yoshikawa[1], **Kazumasa Muramoto**[1] ([1]*Graduate School of Life Science, University of Hyogo*, [2]*Graduate School of Material Science, University of Hyogo*, [3]*School of Life Science, University of Hyogo*, [4]*Graduate School of Pharmaceutical Sciences, Osaka University*, [5]*Institute for Protein Research, Osaka University*)

膜蛋白質／Membrane proteins

3Pos055 脂質・コレステロール・タンパク間の協同性による上皮成長因子受容体の膜近傍ドメイン２量体形成機構
Lipid-cholesterol-protein interaction in the dimerization of juxtamembrane domains of epidermal growth factor receptor
Ryo Maeda[1], Yasushi Sako[1], Takeshi Sato[2] ([1]*Cellular Informatics Lab., RIKEN*, [2]*Kyoto Pharmaceutical Univ.*)

3Pos056 バクテリオルベリン及びその前駆体の膜タンパク質ハロロドプシンへの結合特異性
Binding specificity of bacterioruberin and its precursors to membrane protein halorhodopsin
Fumiya Hattori, Takanori Sasaki (*Grad.Sch.Adv.Math.Sci.,Meiji Univ*)

3Pos057 細胞膜上 Akt の 1 分子イメージングによるシグナル伝達機構の研究
A single molecule imaging approach to understand signal transduction mechanism through Akt on the plasma membrane
Hideaki Yoshimura, Takeaki Ozawa (*Sch. Sci., Univ. Tokyo*)

3Pos058 Triton X-100 により可溶化した Proteorhodopsin の光機能中間体の速度論的解析
Kinetic analysis of photointermediates of Proteorhodopsin solubilized with Triton X-100
Airi Yamamoto[1], Fumio Hayashi[2], Takashi Kikukawa[3,4], Masashi Sonoyama[1,5,6] ([1]*Grad. Sch. Sci. Tech., Gunma Univ.*, [2]*Inst. Anal. Cent., Gunma Univ.*, [3]*Fac. Adv. Life Sci., Hokkaido Univ.*, [4]*GI-CoRE, Hokkaido Univ.*, [5]*GIAR., Gunma Univ.*, [6]*GUCFW., Gunma Univ.*)

3Pos059 1 分子イメージングによる TRPV1 チャネル・脂質間相互作用の時空間動態解析
Spatiotemporal analysis of TRPV1 channel-lipid interaction by single molecule imaging
Yutaro Kuwashima[1,2], Masataka Yanagawa[2], Mitsuhiro Abe[2], Yasushi Sako[2], Ryohei Aoyagi[1], Makoto Arita[1] ([1]*Grad. Sch. Pharm., Univ. Keio*, [2]*Wako Inst., Riken*)

核酸結合蛋白質／Nucleic acid binding proteins

3Pos060 The effect of the distance between the RNA sequences recognized by two RNA-binding domains on the affinity of the MSI1-RNA interaction
Wei Hsun Tu[1,2], Keisuke Kamba[1], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Inst. of Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Energy Sci., Kyoto Univ.*)

3Pos061 Computational insights into DNA binding affinity and its repair activity for photolyase/cryptochrome superfamily
Ryuma Sato[1], Yoshiharu Mori[2], Noriaki Okimoto[1], Makoto Taiji[1] ([1]*RIKEN*, [2]*Kitasato univ*)

3Pos062 The directionality regulation mechanism of serine recombinase
Hsiu-Fang Fan (*NSYSU*)

3Pos063 Single-molecule studies of how polyamines stimulate RecA-mediated recombination
Naciye Esma Tirtom[1], Yang Hsu[2], Hung-Wen Li[1] ([1]*NTU*, [2]*NTNU*)

核酸／Nucleic acid

3Pos064 (3SEA-3) 温度上昇とテラヘルツ光照射は転写反応に異なる影響を及ぼす。
(3SEA-3) Terahertz radiation and temperature increase differently affect transcription by RNA polymerase
Masahiko Imashimizu[1], Masahito Tanaka[1], Hiromichi Hoshina[2], Koh Takeuchi[1] ([1]*AIST*, [2]*RIKEN*)

3Pos065 液体状 DNA の相分離を利用したドメインを持つマイクロゲルカプセルの構築
Construction of hydrogel microcapsules with domain by using phase separation of liquid-like DNA
Yuji Nakashima, Yusuke Sato, Masahiro Takinoue (*Dept. of Comp. Sci., Tokyo Tech.*)

3Pos066 DNA 液滴を用いた液-液相分離による動的システム
Dynamic system by liquid-liquid phase separation using DNA droplet
Nozomi Tsumura, Yusuke Sato, Yuji Nakashima, Masahiro Takinoue (*Tokyo Tech*)

3Pos067 DNA 液滴ポーラスマイクロ構造の形成
Porous microstructure formation of DNA droplet
Tetsuro Sakamoto, Yusuke Sato, Masahiro Takinoue (*Tokyo Institute of Technology*)

3Pos068 DNA ゲルの相転移のシミュレーションモデルと解析
Simulation model and analysis of phase transition of DNA gel
Akihiro Yamamoto, Tetsuro Sakamoto, Yusuke Sato, Masahiro Takinoue (*School of Computing, Tokyo Institute of Technology*)

3Pos069 捩じれストレス下におけるヌクレオソーム DNA 解離および H2A/H2B2 量体脱離の自由エネルギープロファイル
Free energy profiles of unwrapping nucleosomal DNA under torsional stress and eviction of the H2A/H2B dimer
Hisashi Ishida, Hidetoshi Kono (*Institute for Quantum Life Science, QST*)

3Pos070 転写開始複合体における DNA 開裂に関連した DNA・タンパク質間相互作用の検討
DNA-Protein Interaction Related to DNA Opening in Transcription Initiation Complex
Genki Shino, Masahiro Shimizu, Shintaroh Kubo, Toru Niina, Shoji Takada (*Dept. of Biophys., Div. of Bio. Sci., Grad. Sch. of Sci., Univ. of Kyoto*)

水・水和／電解質／Water & Hydration & Electrolyte

3Pos071 Poly(*N*-isopropylacrylamide)のコイル-グロビュール転移に伴う水への溶解度の劇的低下の物理
Physics of drastic decrease in water solubility upon coil-to-globule transition of poly(*N*-isopropylacrylamide)
Masao Inoue, Tomohiko Hayashi, Simon Hikiri, Masahiro Kinoshita (*Inst. Adv. Energ., Kyoto Univ.*)

3Pos072 Water-protein interactions coupled with protein conformational transition
Soichiro Kitazawa[1], Takuro Wakamoto[2], Ryo Kitahara[1] ([1]*College of Pharmaceutical Sciences, Rtisumeikan Univ.*, [2]*Graduate School of Life Sciences, Ritsumeikan Univ.*)

3Pos073 SASA モデルにおける蛋白質間相互作用の過安定化
Over-stabilization of protein-protein interaction in solvent accessible surface area model
Kohei Kuroishi, Dan Parkin, Akira Yodogawa, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

3Pos074 Machine-learning approach for water dynamics
Taku Mizukami[1], Viet Cuong Nguyen[3], Hieu Chi Dam[2] ([1]*JAIST, Materials*, [2]*JAIST, Knowledge*, [3]*HPC.Inc*)

発生・分化／Development & Differentiation

3Pos075 (3SFA-7) 三次元構造モデルから発生過程における細胞機能の理解を試みる
(3SFA-7) Attempt to understand the cellular function during developmental process from 3D structural model
Junpei Kuroda[1,4], Takeshi Itabashi[1,2,3], Takako M. Ichinose[1], Shigeru Kondo[4], **Atsuko H. Iwane**[1,2,3] ([1]*Cell Field Struc., BDR, Riken*, [2]*Grad. sch. Integ. Sci. Life, Hiroshima Univ.*, [3]*Spec. Res. Promot. Group, Grad. Sch. Fronti., Biosci., Osaka Univ.*, [4]*Pattern formation, Grad. Sch. Fronti., Biosci., Osaka Univ.*)

3Pos076 脳形態形成におけるニューロン移動と大脳成長の連成数理モデリング
A coupled mathematical modeling for neuronal migration and cerebral growth in brain morphogenesis
Hironori Takeda[1], Yoshitaka Kameo[1,2,3], Taiji Adachi[1,2,3] ([1]*Grad. Sch. Eng, Kyoto Univ.*, [2]*Inst. Front. Life Med Sci., Kyoto Univ.*, [3]*Grad. Sch. Biostudies, Kyoto Univ*)

3Pos077 Modeling of sea urchin gastrulation based on cytoskeleton imaging
Kaichi Watanabe, Naoaki Sakamoto, Akinori Awazu (*Integrated Sciences for Life in Hiroshima University*)

3Pos078 細胞分裂に関わるキネシン 5 の頭部間協調におけるネック領域の役割の高速一分子観察
High-speed single molecule studies for the role of the neck region on the head-head coordination of mitotic kinesin-5
Taiga Yamada[1], Kohei Matsuzaki[2], Michio Tomishige[2] ([1]*Grad. Sch. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*)

3Pos079 DNA ナノスプリングによる負荷を受けながら運動するキネシン 1 の高速一分子観察
High-speed single-molecule observations of kinesin-1 moving under a load from DNA origami nanospring
Kohei Matsuzaki[1], Mitsuhiro Iwaki[2], Michio Tomishige[1] ([1]*Dept. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*, [2]*BDR, Riken*)

3Pos080 Kinetic parameters and reaction scheme of high and low activity mutants of Serratia marcescens chitinase A
Akasit Visootsat[1,2], Paul Vignon[3], Akihiko Nakamura[1,2], Takayuki Uchihashi[4,5], Hiroki Watanabe[4,5], Ryota Iino[1,2] ([1]*IMS*, [2]*SOKENDAI*, [3]*ParisTECH*, [4]*Nagoya University*, [5]*ExCELLS*)

3Pos081 高速原子間力顕微鏡により観察された微小管上の外腕ダイニン
High-speed atomic force microscopy on outer dynein arms aligned on microtubules
Kenta Ishibashi[1,2], Kazuhiro Oiwa[2,3,4] ([1]*Osaka Univ.*, [2]*NICT-CiNet*, [3]*Advanced ICT Research Institute*, [4]*University of Hyogo*)

3Pos082 Development of novel Photochromic inhibitors for kinesin Eg5 which form multiple isomerization states utilizing azobenzene and spiropyran
Islam Md Alrazi, Kei Sadakane, Shinsaku Maruta (*Department of Bioinformatics, Graduate School of Engineering, Soka University, Hachioji, Tokyo , Japan*)

3Pos083 回転子変異体を用いたべん毛モータースイッチ機構の解析
Analysis of the bacterial flagellar switch using mutant rotor components
Mai Kato[1], Tsubasa Ishida[1], Myu Yoshida[2], Yoshisyuki Sowa[1,2,3] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.,*, [2]*Dept. Frontier Biosci., Hosei Univ*, [3]*Micro-nano Tech., Hosei Univ*)

3Pos084 細菌べん毛モーターの回転を支える回転軸-軸受間相互作用の解析
Analysis of shaft-bearing interactions that support the smooth rotation of bacterial flagellar motors
Yumi Kumazaki[1], Tsubasa Ishida[1], Myu Yosida[2], Yoshiyuki Sowa[1,2,3] ([1]*Grad. Sch. Sci. & Eng, Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Micro-nano Tech., Hosei Univ.*)

3Pos085 Controlling the rotation speed of the bacterial flagellar motor with light-driven rhodopsin
So Hasegawa[1], Rei Abe-Yoshizumi[2], Keiichi Inoue[3], Hideki Kandori[2], Yoshiyuki Sowa[4] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ*, [2]*Grad. Sch. Eng., Nagoya Inst. Tech*, [3]*Inst. Solid State Phys., Univ. Tokyo*, [4]*Dept. Frontier Biosci., Hosei Univ*)

3Pos086 *Paenibacillus* sp. TCA20 と大腸菌に由来するべん毛モーターキメラ固定子のイオン選択性
Ion specificity of chimeric stator proteins between *Paenibacillus* sp. TCA20 MotB1 and *Escherichia coli* MotB
Sakura Onoe[1], Myu Yoshida[2], Masahiro Ito[3], Yoshiyuki Sowa[1,2,4] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Grad. Sch. Life Sci. Toyo Univ.*, [4]*RC. Micro-nano Tech., Hosei Univ.*)

3Pos087 DNA オリガミを用いた野生型と変異体キネシン二分子による協調運動の観察
Cooperative transport by wild-type and mutant kinesin motors as studied by using programmable DNA origami
Shu Takano[1], Ryosuke Masuda[3], Kohei Matsuzaki[2], Mitsuhiro Iwaki[4], Michio Tomishige[2] ([1]*Grad. Sch. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*, [3]*Dept. Appl. Phys., Grad. Sch. Eng., Univ. Tokyo*, [4]*BDR, Riken*)

3Pos088 INHIBITION OF MITOTIC KINESIN EG5 BY KOLAFLAVANONE
Tomisin Ogunwa[3], Kei Sadakane[1], Ayodele O. Kolawole[4], Olusola O. Elekofehinti[4], Afolabi C. Akinmoladun[4], Olaposi I. Omotuyi[5], Takayuki Miyanishi[3], **Shinsaku Maruta**[1,2] ([1]*Sci. & Engin., Soka University*, [2]*Grad. Sch. Engin., Soka Unive.*, [3]*Grad. Sch. Fisheries and Environmental Sci, Nagasaki Unive.*, [4]*Dept. of Biochem., The Federal Unive. of Tech.*, [5]*Centre for Biocomputing and Drug Design, Adekunle Ajasin Unive.*)

3Pos089 F_1-ATPase の軸とシリンダーの結合寿命の測定
Single-molecule pull-out manipulation of the shaft of the rotary motor F_1-ATPase
Tatsuya Naito[1], Tomoko Masaike[2], Daisuke Nakane[1], Mitsuhiro Sugawa[3], Takayuki Nishizaka[1] ([1]*Dept. Phys., Gakushuin Univ.*, [2]*Dept. Appl. Biol. Sci., Tokyo Univ. Sci.*, [3]*Grad school of arts and sciences, Univ. of Tokyo,*)

3Pos090 Does giraffe kinesin move faster than mouse?
Taketoshi Kambara[1], Yasushi Okada[1,2] ([1]*RIKEN BDR*, [2]*U. of Tokyo, Grad. Sci.*)

3Pos091 Microsecond-resolved observation of F_1-ATPase conformational changes by single molecular fluorescence spectroscopy
Hiroki Senmaru[1], Hiroyuki Oikawa[2], Mitsuhiro Sugawa[3], Satoshi Takahashi[2] ([1]*Tohoku University Graduate school of life sciences*, [2]*Tohoku university IMRAM*, [3]*Tokyo university graduate school of Arts and Sciences*)

3Pos092 遺伝子工学的に人工的に設計したモータタンパク質収縮ネットワークの性能向上
Improvement of a genetically-engineered microtubule contractile protein network
Zhao Du[1], Takahiro Nitta[3], Yingzhe Wang[2], Keisuke Morishima[2], Yuichi Hiratsuka[1] ([1]*JAIST, Sch. of Mat. Sci.*, [2]*Osaka Univ., Grad. Sch. of Eng., Dep. Mech. Eng.*, [3]*Gifu Univ., Grad. Sch. of Eng., Dep. of EECE*)

3Pos093 暗視野顕微鏡を用いた微小管混雑時における細胞質ダイニンのステップの高時間分解能観察
Cytoplasmic dynein stepping on crowded microtubules resolved using dark-field imaging with high spatio-temporal resolution
Yusuke Kumagai[1], Keitaro Shibata[2], Ken'ya Furuta[2], Hajime Honda[1], Hiroaki Kojima[2] ([1]*Dep. Bioeng., Nagaoka Univ.*, [2]*Adv. ICT Res. Ins., NICT*)

3Pos094 共通祖先型 F_1-ATPase の一分子回転解析
Reconstruction and Characterization of Ancestral F_1-ATPase
Nanako Nakama[1], Hiroshi Ueno[1], Ryutaro Furukawa[2], Ryohei Kobayashi[1], Ryo Watanabe[1], Satoshi Akanuma[2], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*Facul. Human Sci., Univ. Waseda*)

3Pos095 クラミドモナス軸糸ダイニン集合体の外部負荷に対する応答を測定する
Measuring mechanical responses of Chlamydomonas axonemal dynein arrays to external load
Misaki Sagawa[1], Akane Furuta[2], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2], Ken'ya Furuta[2] ([1]*Grad. Sch. of Life Science, Univ. Hyogo*, [2]*Adv. ICT Res. Inst., NICT*)

細胞生物的課題／Cell biology

3Pos096 (3SHA-4) 過渡的に形成される GPCR ダイマーの研究：細胞内蛍光 1 分子観察によるアプローチ
(3SHA-4) Examining the transiently formed GPCR dimer: an approach by single fluorescent molecule observation in living cells
Rinshi Kasai (*Inst. Front. Life. Med. Sci., Kyoto Univ.*)

3Pos097 微小管結合蛋白質を介したアクチンフィラメントと微小管との束化が細胞突起に与える影響
The role of microtubule-associated protein mediated bundle formation between actin filaments and microtubule on cell process formation
Chihiro Doki[1], Masahiro Kuragano[1], Kohei Nishida[1], Shoma Saito[1], Susumu Kotani[2], Kiyotaka Tokuraku[1] ([1]*Grad.Sch.Eng.,Muroran Inst.Tech.*, [2]*Fac. Sci., Kanagawa Univ*)

3Pos098 デスミンとアクチンを包含する液滴のそれらの集合により誘発される変形
Deformations of droplets containing desmin and actin caused by their assembly
Yoshiya Miyasaka, Keigo Murakami, Kuniyuki Hatori (*Grad. Sch. of Sci. and Eng., Yamagata Univ.*)

3Pos099 S1P 修飾弾性率可変ゲルを用いた Muse 細胞ホーミング及び力学場応答性の解析
Homing and mechano-response of Muse cells analyzed on S1P-modified hydrogel with tunable elasticity
Lei Guo[1], Yukie Tsuji[2], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

3Pos100 細胞の突起形成における MAP4 の局在と機能
Localization and function of microtubule-associated protein (MAP) 4 in cell protrusion formation
Kohei Nishida[1], Masahiro Kuragano[1], Chihiro Doki[1], Susumu Kotani[2], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Eng., Muroran Inst. Tech.*, [2]*Fac. Sci., Kanagawa Univ.*)

3Pos101 F-actin に沿った Fimbrin の協同的クラスター形成の方向性
Direction of the cooperative cluster formation of fimbrin along actin filaments
Naoki Hosokawa[1], Masahiro Kuragano[1], Keitaro Shibata[2], Taro Q.P. Uyeda[3], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Eng., Muroran Inst. Tech.*, [2]*NICT*, [3]*Dep. of Physics, Fac. Sci. Engin., Waseda Univ.*)

3Pos102 非凍結温度において氷結合タンパク質は線虫の低温耐性を改善する
Ice-Binding Proteins Improves the Survival Rate of *Caenorhabditis elegans* at Non-freezing Temperature
Masahiro Kuramochi[1,2,3], Geikaku Tou[1], Chiaki Takanashi[1], Motomichi Doi[3], Kazuhiro Mio[2], Sakae Tsuda[4], C. Yuji Sasaki[1,2] ([1]*University of Tokyo*, [2]*AIST-UTokyo OIL*, [3]*Biomedical R.I., AIST*, [4]*Bioproduct R.I., AIST*)

3Pos103 フェムト秒レーザー誘起衝撃力と反射干渉顕微鏡による細胞接着強度の定量評価手法の確立
Quantitative evaluation of cell adhesion strength by reflection interference contrast microscopy combined with femtosecond laser impulse
Yukiko Yoshimura, Sohei Yamada, Yoichiroh Hosokawa, Ryohei Yasukuni, Kazunori Okano (*Division of Materials Science, Nara Institute of Science and Technology*)

3Pos104 アクチンネットワーク上におけるアクチン結合タンパク質の局在形成における自律的制御機構
Self-regulatory mechanisms for the segregation of actin binding proteins on actin network
Yosuke Yamazaki, Taro QP Uyeda (*Dep. Phys., Waseda Univ.*)

3Pos105 iPS 細胞は最適弾性領域に移動し、増殖促進と高質な幹細胞性保持を示す
iPS cells show mechanotactic accumulation, higher proliferation and expression of stemness marker in optimal region of matrix elasticity
Mengfan Wang[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu. Univ.*, [2]*IMCE, Kyushu Univ.*)

3Pos106 分化フラストレート MSC における APC 発現調節の上流機構の検証
Investigation of upstream regulatory factors of APC expression in the MSCs in frustrated differentiation
Misaki Kaneshiro[1], Thasaneeya Kuboki[2], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

3Pos107 Ca^{2+} 存在下/非存在下における mbo1（後退運動変異株）の鞭毛波形
The flagellar waveforms of *mbo1*, a mutant moving backward only, in the presence/absence of Ca^{2+}
Hitoshi Sakakibara, Hiroaki Kojima, Kazuhiro Oiwa (*Adv. ICT Res. Inst., NICT*)

3Pos108 アクチン結合タンパク質 Rng2 がアクトミオシン in vitro 運動を協同的に阻害するメカニズム
The mechanism of cooperative inhibition of actomyosin movement in vitro by the actin binding protein Rng2
Yuuki Hayakawa[1], **Yosuke Kakuta**[1], Ngo Kien X.[2], Noriyuki Kodera[2], Taro QP Uyeda[1] ([1]*Department of Physics, Faculty of Advanced Science and Engineering, Waseda University*, [2]*Bio-AFM Res. Ctr., Kanazawa Univ.*)

3Pos109 二種の異なるアクチン結合タンパク質のアクチンへの結合が相互排他的かまたは協調的かを評価する観察系の構築
Establishment of observation system to evaluate whether two different actin binding proteins bind to actin mutually or accommodative
Tenji Yumoto, Taro QP Uyeda (*Dept. Physics, Waseda Univ.*)

3Pos110 動的な誘引物質濃度勾配における好中球様 HL60 細胞の運動方向決定
Decision making of migratory direction of neutrophil-like HL60 cells in dynamical chemoattractant gradient
Motohiko Ishida[1], Akihiko Nakajima[2,3], Satoshi Sawai[1,3] ([1]*Dept. Basic Sci., Grad. Sch. of Arts & Sci., Univ. of Tokyo, Japan*, [2]*Dept. General Systems Studies, Grad. Sch. of Arts & Sci., Univ. of Tokyo, Japan*, [3]*Comp. Sys. Biol. Cent., Grad. Sch. of Arts & Sci., Univ. of Tokyo, Japan*)

3Pos111 タウ–微小管相互作用の等温滴定熱測定
Isothermal titration calorimetry of tau-microtubule interaction
Junta Kashima, Hiroshi Sakamoto, Junichi Taira, Hideyuki Komatsu (*Biosci. Bioinf., Kyushu Inst. Tech.*)

3Pos112 Rhodamine-phalloidin と Lifeact-GFP のアクチン結合の相互排他性
Mutual Exclusion of Actin Binding between Rhodamine-phalloidin and Lifeact-GFP
Saku Kijima[2], **Yuuya Aoki**[1], Taro QP Uyeda[1] ([1]*Dept. Physics, Waseda Univ*, [2]*Biopro. Res. Inst., AIST*)

3Pos113 RhPh 染色したアクチンフィラメントの 3 つの蛍光の明滅パターンの解析
Analysis of three distinct blinking patterns of RhPh fluorescence along actin filaments
Kazunori Ono, Ryuichi Kaneda, **Syunsuke Ando**, Koki Arai, Yosuke Yamazaki, Taro QP Uyeda (*Dept. Physics, Waseda Univ*)

3Pos114 らせん型細菌スピロヘータの遊泳の力と速度の関係
Force-velocity relationship of the spirochete *Leptospira* swimming
Keigo Abe[1], Kyosuke Takeba[2], Shuichi Nakamura[1] ([1]*Grad.Sch.Eng., Tohoku Univ.*, [2]*Life and Env.Sci., Tsukuba Univ.*)

生体膜・人工膜／Biological & Artificial membrane: Dynamics

3Pos115 Effect of sucrose on the diffusion of proteins tethered in a glass-supported lipid bilayer
Hiromitsu Hariu (*Saitama Univ.*)

3Pos116 分子ツールとしての電位依存性ホスファターゼの改良
Improvement of voltage-sensing phosphatase as a molecular tool of phosphoinositide depletion in living cells
Akira Kawanabe[1,2], Natsuki Mizutani[2], Tomoko Yonezawa[2], Yasushi Okamura[2] ([1]*Fac. Med., Kagawa Univ.*, [2]*Grad. Sch. Med., Osaka Univ.*)

3Pos117 油中水滴接触膜張力の定量的操作法とチャネル研究への応用
Manipulation and quantitative evaluation of membrane tension during single-channel current recordings in the contact bubble bilayer
Masayuki Iwamoto[1], Shigetoshi Oiki[2] ([1]*Dept. Mol. Neurosci., Univ. Fukui. Facul. Med. Sci.*, [2]*Biomed. Imaging Res. Center, Univ. Fukui*)

3Pos118 細菌機械受容チャネル MscL の脂質膜環境変化に対する応答のシミュレーション研究
Computational Study Focusing on the Response to Changes of Membrane Environment in Gating of the Bacterial Mechanosensitive Channel MscL
Yasuyuki Sawada[1], Ken'ichi Hashimoto[2], Hisashi Kawasaki[2], Masahiro Sokabe[3] ([1]*Dept. Nutrition, Nagoya Univ. Eco.*, [2]*Biotech. Res. Ctr., Tokyo Univ.*, [3]*Mechanobiology Lab, Nagoya Univ. Grad. Sch. Med.*)

3Pos119 A gold nano-electrode for single channel detection
Toru Ide[1,2], Minako Hirano[2], Kota Kaneko[1], Huimin Ma[1] ([1]*Fac. Engn. Okayama Univ.*, [2]*Photo-Bio. GPI*)

3Pos120 全自動パッチクランプシステムによる、アダプティブコントロールを用いた正確な 50%不活性化状態制御実験の実現
Adaptive voltage control ensures the precise half inactivation application of voltage gated channels on automated patch clamp system
Kazuya Tsurudome (*Sophion Bioscience K.K.*)

3Pos121 人工イオンチャネルの分子動力学シミュレーション
All-atom molecular dynamics simulations of artificial ion channels
Takahiro Osamura[1], Toru Ekimoto[1], Tsutomu Yamane[1], Takahiro Muraoka[2], Kazushi Kinbara[3], Mitsunori Ikeguchi[1,4] ([1]*Grad. Sch. Med Life Sci., Yokohama City Univ.*, [2]*Grad. Sch. Global Innov., Tokyo Univ. of Agri. and Tech.*, [3]*Grad. Sch. Life Sci. and Tech., Tokyo Tech*, [4]*Med. Sci. Innov. Hub., Riken*)

3Pos122 筋小胞体カルシウムポンプの Ca^{2+}結合に及ぼす界面活性剤の効果
Effect of solubilization with a detergent on sarcoplasmic reticulum Ca^{2+}-ATPase
Takashi Daiho, Stefania Danko, Kazuo Yamasaki, Satoshi Yasuda, Hiroshi Suzuki (*Asahikawa Medical Univ.*)

3Pos123 プロトニック有機電極によるミトコンドリアの ATP 合成操作
Control of mitochondrial ATP synthesis with a protonic biotransducer
Momoka Takahashi[1], Mingyin Cui[2], Hiroko Kashiwagi[1], Takeo Miyake[2], Yoshihiro Ohta[1] ([1]*Grad. Sch. Biotech., TUAT*, [2]*Grad. Sch. Info., Univ. Waseda*)

3Pos124 Toward the construction of DNA origami artificial channel with selective transport function
Shoji Iwabuchi[1], Ibuki Kawamata[1], Yuki Suzuki[2], Satoshi Murata[1], M. Shinichiro Nomura[1] ([1]*Grad. Eng., Univ. Tohoku*, [2]*FRIS, Univ. Tohoku*)

3Pos125 上皮成長因子受容体クラスターによる EGF シグナル伝達の調節
Regulation of downstream signaling by clusters of epidermal growth factor receptor
Michio Hiroshima[1,2], Nario Tomishige[3], Masahiro Ueda[1], Toshihide Kobayashi[3], Yasushi Sako[2] ([1]*RIKEN BDR*, [2]*RIKEN CPR*, [3]*Univ. of Strasbourg*)

3Pos126 p52Shc は時間依存的に Grb2 のシグナル伝達ダイナミクスを制御する
p52Shc regulates Grb2 signaling dynamics in a time dependent manner after cell stimulation
Ryo Yoshizawa[1], Nobuhisa Umeki[2], Masataka Yanagawa[2], Masayuki Murata[1], Yasushi Sako[2] ([1]*Grad.sch.arts and ahi., the univ. Tokyo*, [2]*Wako Inst., Riken*)

3Pos127 Analysis of electrostatic interaction of acidic glycolipid with transmembrane peptide of insulin receptor
Yuka Nimura[1], Kazuya Kabayama[1,2,3], Yuya Asahina[4], Shinya Hanashima[1], Hironobu Hojo[4], Michio Murata[1], Koichi Fukase[1,2,3] ([1]*Grad. Sch. of Sci., Osaka Univ.*, [2]*MS-CORE, PRC, Grad. Sch. of Sci., Osaka Univ.*, [3]*Inst. for Radiation Sciences, Osaka Univ.*, [4]*Inst. for Protein Research, Osaka Univ.*)

3Pos128 細胞膜断片ブレブを用いたモデル生体膜への膜タンパク質再構成
Direct reconstitution of membrane proteins from cell membrane blebs into a model biological membrane
Rurika Nagai[1], Yasushi Tanimoto[2], Rinshi Kasai[3], Kenichi Suzuki[4,5], Fumio Hayashi[6], Kenichi Morigaki[1,2] ([1]*Grad. Sch. Agr., Univ. Kobe*, [2]*Biosignal Research Center, Univ Kobe*, [3]*Institute for Frontier Life and Medical Sciences, Univ Kyoto*, [4]*G-chain, Univ. Gifu*, [5]*Grad of Nat. Sci and Tech., Univ Gifu*, [6]*Grad. Sch. Scie, Univ. Kobe*)

神経回路・脳の情報処理／Neuronal circuit & Information processing

3Pos129 アガロース微細構造を用いた海馬細胞から伸長する神経突起の相互作用の解析
Interactions of neurites elongated from isolated hippocampal cells in agarose width-length-controlled microchannels
Yuhei Tanaka[1], Shota Aoki[1], Haruki Watanabe[2], Kenji Shimoda[2], Akihiro Hattori[3], Masao Odaka[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

3Pos130 Implementation of automated driving by deep reinforcement learning on high definition simulator
Shunsuke Isomura[1], **Hideo Mukai**[1,2] ([1]*Comp. Sci. Prog., Grad. Sch. Sci & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci & Tech., Meiji Univ.*)

3Pos131 単一の神経突起における伸長特性を測定するためのスポット吸収マイクロニードルを用いた μm 単位の精度のアガロース微細加工技術
Precise μm agarose microfabrication technology with spot absorption microneedle for single neurite elongation property measurement
Haruki Watanabe[1], Yuhei Tanaka[2], Shota Aoki[2], Kenji Simoda[1], Takahito Kikuchi[2], Akihiro Hattori[3,4], Masao Odaka[3,4], Kenji Yasuda[1,2,3,4] ([1]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*, [4]*WASEDA Biosci. Res. Ins. in Singapore (WABIOS)*)

3Pos132 畳み込みニューラルネットワークを用いた脳波解析手法の実装
Implementation of EEG analysis method with Convolutional Neural Networks
Hiroaki Takao[1], Hideo Mukai[1,2] ([1]*Comp. Sci. Prog., Grad Sch. Sci & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci. & Tech., Meiji Univ.*)

光生物学：視覚・光受容／Photobiology: Vision & Photoreception

3Pos133 リガンド非結合時のオプシンは稀に光活性化したロドプシンと同等の活性を示す
Apo-opsin exists in equilibrium between a predominantly inactive and a rare highly active state
Shinya Sato[1,2], Beata Jastrzebska[3], Andreas Engel[3], Krzysztof Palczewski[3,4], Vladimir J. Kefalov[1] ([1]*DOVS, Washington Univ.*, [2]*Grad. Sch. Biostudies., Kyoto Univ.*, [3]*Case Western Reserve Univ.*, [4]*UC Irvine*)

3Pos134 天然のバクテリアを用いたヘリオロドプシンの機能研究
Function study of heliorhodopsin using native bacteria
Ai Muto, Rei Abe-Yoshizumi, Hideki Kandori (*Nagoya Inst. Tech.*)

3Pos135 メラノプシンの光活性化機構
Photoactivation process of Melanopsin
Masami Kugo[1], Takahiro Yamashita[1], Yoshinori Shichida[2], Yasushi Imamoto[1] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Ritsumeikan Univ.*)

3Pos136 アミノ酸置換による脊椎動物ロドプシンのバイステーブル特性化
Construction of vertebrate rhodopsin with bistable property by a single mutation
Kazumi Sakai[1], Yoshinori Shichida[2], Takahiro Yamashita[1] ([1]*Graduate School of Science, Kyoto University*, [2]*Research Organization for Science and Technology, Ritsumeikan University*)

3Pos137 高角 X 線散乱法による光活性化ロドプシンの活性構造安定化メカニズムの解析
Stabilization Mechanism of Active Conformation of Photoactivated Rhodopsin Studied by High-Angle X-Ray Scattering
Yasushi Imamoto[1], Keiichi Kojima[2], Toshihiko Oka[3], Ryo Maeda[4], Yoshinori Shichida[5] ([1]*Grad. Sch. Sci., Kyoto Univ.*, [2]*Grad. Sch. Med. Dent. Pharm. Sci., Okayama Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*, [4]*RIKEN*, [5]*Ritsumeikan Univ.*)

3Pos138 シアノバクテリオクロム型光受容体のシステイン残基の着脱反応におけるプロトンの役割
Protochromic Absorption Changes in Two-Cys Photocycle of the Blue/Orange Cyanobacteriochrome
Teppei Sato[1,2], Takashi Kikukawa[3], Risako Miyoshi[4], Kosuke Kajimoto[4], Chinatsu Yonekawa[1], Tomotsumi Fujisawa[4], Masashi Unno[4], Toshihiko Eki[1], **Yuu Hirose**[1] ([1]*Toyohashi Univ. of Tech.*, [2]*Nagoya Univ.*, [3]*Hokkaido Univ.*, [4]*Saga Univ.*)

3Pos139 パターン化モデル膜を用いたロドプシンリン酸化とアレスチン結合の解析
Rhodopsin phosphorylation and arrestin binding studied in a patterned model membrane
Fuko Kueda[1], Yasushi Tanimoto[2], Fumio Hayashi[3], Kenichi Morigaki[2,4] ([1]*Fac. Agr., Univ. Kobe*, [2]*Biosignal Research Center, Univ. Kobe*, [3]*Grad. Sch. Scie., Univ. Kobe*, [4]*Grad. Sch. Agr., Univ. Kobe*)

3Pos140 長波長光感受性視物質の塩化物イオン結合における Gln114 の役割
Role of Gln114 in chloride binding of long-wavelength-sensitive visual pigment
Kota Katayama[1], Shunta Nakamura[1], Takuma Sasaki[1], Hiroo Imai[2], Hideki Kandori[1] ([1]*Grad.Sch.Eng., Nagoya Inst. Tech*, [2]*Orimate Res. Inst., Kyoto Univ*)

3Pos141 Comparative Study for Anion Transport Activity of Anion Channelrhodopsins by Using a Simple pH Electrode Method
Chihiro Kikuchi (*Grad. Sch. Life Sci.*)

光生物学：光合成／Photobiology: Photosynthesis

3Pos142 (3SDA-3) 生体組織への応用が期待される光感度の高いチャネルロドプシン
(3SDA-3) Novel optogenetics tool: A light-gated cation channel with high-reactivity to weak light
Shoko Hososhima[1], Shunta Shigemura[1], Hideki Kandori[1], Satoshi Tsunoda[1,2] ([1]*Nagoya Institute of Technology*, [2]*JST, PRESTO*)

3Pos143 光依存的にホモオリゴマー化する植物クリプトクロム 2 の分子特性
Molecular properties of light-dependent homo-oligomerizing *At* CRY2
Kazuya Agata[1], Daichi Yamada[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech., Dept. Life Sci. Appl. Chem.*, [2]*Univ. Hyogo, Dept. Life Sci.*)

3Pos144 光ジッパーを用いた bZIP 型転写因子の標的配列認識の解析
Analyses of the target sequence recognition of a bZIP factor, using a light-activatable Photozipper
Osamu Hisatomi, Samu Tateyama, Itsuki Kobayashi (*Grad. Sch. of Sci., Osaka Univ.*)

3Pos145 光制御型 bZIP 転写因子（フォトジッパー）の β シート疎水面の役割
Hydrophobic residues on the β-sheet of a light-activatable bZIP factor, Photozipper
Hiroto Nakajima, Itsuki Kobayashi, Osamu Hisatomi (*Grad.Sch.of Sci.,Osaka Univ.*)

3Pos146 高速 AFM による DNA 結合光受容タンパク質 Photozipper の 1 分子動態イメージング
Single molecular dynamics imaging of DNA binding photoreceptor protein, Photozipper, by high-speed AFM
Kento Nomura[1], **Hayato Yamashita**[1], Osamu Hisatomi[2], Masayuki Abe[1] ([1]*Grad. Sch. of Eng. Sci., Osaka Univ.*, [2]*Grad. Sch. of Sci., Osaka Univ.*)

3Pos147 Theoretical study on molecular mechanism of a light-driven ion transport of Halorhodopsin from Natronomonas pharaonis
Ryo Oyama, Taisuke Hasegawa, Shigehiko Hayashi (*Grad. Sch. Sci., Univ. Kyoto*)

ゲノム生物学／Genome biology

3Pos148 ウニ初期胚核内構造の発生に伴う動的変化とその細胞特異性
Dynamic and cell specific changes in intranuclear chromosomal
Yuhei Yasui (*Integrated science for life, Hiroshima Univ*)

3Pos149 ヌクレオソーム排他的ループ非形成型インスレーター配列（NENLIS）によるインスレーター活性のゲノムワイド解析
Genome-wide analysis of insulator activity by nucleosome exclusive non-looping insulator sequence (NENLIS)
Yudai Hirose[1], Yuki Matsushima[1], Naoaki Sakamoto[2], Akinori Awazu[2] ([1]*Grad. Sch. Sci., Univ. Hiroshima*, [2]*Grad. Sch. Integrated Sci., Univ. Hiroshima*)

3Pos150 X 染色体不活性化を誘導する染色体動態
The dynamics of chromosomes that trigger X chromosome inactivation
Tetsushi Komoto, Hiraku Nishimori, Akinori Awazu (*Integrated Science for Life in Hiroshima university*)

生命情報科学／Bioinformatics

3Pos151 Machine learning models for predicting ligand-binding sites using residue-wise features
Masafumi Shionyu, Atsushi Hijikata (*Fac. Biosci., Nagahama Inst. Bio-Sci. Tech.*)

3Pos152 モノクローナル抗体の詳細と提供情報のデータベース
Database for information of acquirable monoclonal antibody
Hirofumi Suzuki[1], Mika Kaneko[2], Yukinari Kato[2], Kei Yura[1,3] ([1]*Dept. Life Sci. & Med. Bio., Waseda Univ.*, [2]*Grad. Sch. of Med., Tohoku Univ.*, [3]*Sim. Info. Bio., Ochanomizu Univ.*)

3Pos153 強力な順方向遺伝学による生物システム解析 -Vibrio alginolyticus の走性への応用
Biological system analysis by a strong forward genetics -An application to a taxis in a strain Vibrio alginolyticus
Kunio Ihara[1], Kazuma Uesaka[1], Noriko Nishioka[2], Seiji Kojima[2], Michio Homma[2] ([1]*Nagoya University Cent.Gene Res.*, [2]*Nagoya University Grad. School Sci.*)

3Pos154 時点数の少ないトランスクリプトームデータからのネットワーク推定に適した遺伝子グループ化法
Gene grouping strategy for network inference from a small time-series transcriptome data
Kiyohiro Maeda (*Fujifilm Corporation*)

3Pos155 生物発光タンパク質およびルシフェリンの獲得進化解析
Evolutionary Analysis of Luciferase, Photoprotein and Luciferin
Misato Funahashi[1], Hirofumi Suzuki[2], **Kei Yura**[1,2] ([1]*Grad. Schl Hum. Sci., Ochanomizu Univ.*, [2]*Schl Adv. Sci. Engng., Waseda Univ.*)

数理生物学／Mathematical biology

3Pos156 二倍体遺伝子発現制御系における集合的メンデル遺伝
Group Mendelian Dominance in Diploid Gene Regulatory Network
Kenji Okubo, Kunihiko Kaneko (*Dep. of Basic Sci., Univ. Tokyo*)

3Pos157 不正確な素子から機能的なネットワークをつくる方法
Cooperative architecture for functional network from sloppy gene expression dynamics
Masayo Inoue[1], Kunihiko Kaneko[2] ([1]*IMS, Meiji Univ.*, [2]*Univ. of Tokyo*)

3Pos158 混雑下のナノ～マイクロマシン集団：内部状態と環境の相互干渉
Nano/Micro-machines in the Crowd: Interplay between the Internal State and Surroundings
Yuichi Togashi[1,2] ([1]*RIKEN BDR*, [2]*Grad. Sch. Integ. Sci. Life, Hiroshima Univ.*)

3Pos159 3D phase field simulation for macropinocytosis of amoeboid cells
Nen Saito[1], Satoshi Sawai[2] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Grad. Sch. Arts Sci., Univ. Tokyo*)

3Pos160 Geometric feature extraction from some gene expression pattern for prediction of atopic dermatitis patients
Takuya Hasebe[1], Masahiro Sugimoto[2], Takanori Sasaki[1] ([1]*Grad. Sch. Adv. Math. Sci., Meiji Univ.*, [2]*RDCMIT, Tokyo Med. Univ.*)

3Pos161 増殖系と隠れマルコフモデルの対応に基づく学習の考察
A Study on Learning in Growing Population on the Basis of Hidden Markov Model
So Nakashima[1], Tetsuya J. Kobayashi[2] ([1]*grad. school of IS&T, UTokyo*, [2]*IIS, UTokyo*)

3Pos162 脳神経系の動的ネットワークモデルにおける自発的階層構造形成
Spontaneous hierarchical structure formation in dynamic network model of cerebral nervous system
Amika Ohara, Hiraku Nishimori, Akinori Awazu (*Dept. of math. and life sci. Hiroshima univ.*)

3Pos163 Fitness response relation of a multitype age-structured population dynamics
Yuki Sughiyama[1], So Nakashima[2], Tetsuya Koabayasi[1] ([1]*IIS, The University of Tokyo*, [2]*Department of Mathematical Informatics, The University of Tokyo*)

3Pos164 Simulation and regulation of E.coli which has autonomous diversification ability
Eriko Nakagawa[1], Shotaro Ayukawa[2], Daisuke Kiga[1] ([1]*Department of Electrical Engineering and Bioscience, Waseda University*, [2]*Waseda Research Institute for Science and Engineering, Waseda University*)

3Pos165 Probability landscape of coupled epigenetic and genetic network with eddy-like probability currents
Bhaswati Bhattacharyya, Masaki Sasai (*Department of Applied Physics, Nagoya University*)

3Pos166 ドロップアウトを適用したニューラルネットワークアルゴリズムによる大腸菌の遺伝子制御ネットワークの推定
Inference of gene regulatory network of E.coli by neural network algorithm applied dropout
Yusuke Mizukoshi[1], Masahiro Sugimoto[2], Takanori Sasaki[3] ([1]*Grad. Sch. Adv. Math. Sci., Univ.Meiji*, [2]*RDCMIT, Univ.Tokyo Med*, [3]*Grad. Sch. Adv. Math. Sci., Univ.Meiji*)

非平衡・発生リズム／Nonequilibrium state & Biological rhythm

3Pos167 Active Nematic Sperms in Vivo Mouse
Tsuyoshi Hirashima[1], Kyogo Kawaguchi[2], Takuya Omotehara[3], Masahiro Itoh[3], Kenta Ishimoto[4], Michiyuki Matsuda[1,5] ([1]*Grad Sch Med, Kyoto Univ*, [2]*RIKEN*, [3]*Tokyo Medical University*, [4]*Grad Sch Math Sci, The Univ of Tokyo*, [5]*Grad Sch Biostudies, Kyoto Univ*)

3Pos168 メチルセルロースとの相分離が誘起する重合過程アクチン線維の紡錘形液晶ドメイン自発形成
Emergence of spindle-shaped namatic domains of filamentous actin during polymerization induced by phase-separation from methylcellulose
Masahito Hayashi[1], Tomoyuki Kaneko[1], Kingo Takiguchi[2] ([1]*LaRC, Frontier Biosci., Hosei Univ.*, [2]*Dept. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

3Pos169 Traveling band formation of a mutant *Dictyostelium* cell population induced by contact following of locomotion
Masayuki Hayakawa[1], Tetsuya Hiraiwa[2], Yuko Wada[1], Hidekazu Kuwayama[3], Tatsuo Shibata[1] ([1]*Riken BDR*, [2]*Dept. of Phys, Univ. of Tokyo*, [3]*Faculty of Life and Env. Sci., Univ. of Tsukuba*)

3Pos170 高速 AFM による KaiC、KaiB、SasA 間相互作用のリン酸化状態依存性観察
High-Speed AFM observation of phosphorylation state-dependent interactions between KaiC, KaiB and SasA
Kenta Ueda[1], Tetsuya Mori[2], Shogo Sugiyama[3], Takayuki Uchihashi[1], Carl H. Johnson[2] ([1]*Dept. of Sci, Nagoya Univ*, [2]*UnivDept.of Biol.Sci., Vanderbilt Univ*, [3]*Dept. of Phys, Kanazawa*)

3Pos171 Individual cyanobacterial circadian rhythms under chilly conditions
Hiroshi Ito[1], Hinako Maruayama[1], Irina Mihalcescu[2] ([1]*Grad. Sch. Design., Kyushu Univ.*, [2]*LIPhy, Universite Grenoble Alpes*)

計測／Measurements

3Pos172 多電極システムを用いた心毒性検査のためのハイスループットチャンバーの改良
Improvement of high-throughput chamber for cardio-toxicity testing with multi-electrode array system
Naoki Tadokoro, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

3Pos173 HPD による広視野蛍光 1 分子偏光検出
Wide-field single-molecule fluorescence polarization detection by hybrid photo-detectors (HPDs)
Atsuhito Fukasawa[1], Gaku Nakano[1], Minako Hirano[2], Toru Ide[3], Hiroaki Yokota[2] ([1]*Hamamatsu Photonics K.K.*, [2]*Grad. Sch. Creation Photon Indust.*, [3]*Grad. Sch. Interdisciplinary Sci. and Engineering in Health Sys.*)

3Pos174 蛍光タンパク質の赤外スペクトル測定ー発色団部位の選択的な観測ー
IR spectra of fluorescent proteins -selective measurement of chromospheres-
Hirona Takahashi, Makoto Sakai (*faculty of Science, Okayama University of Science*)

3Pos175 蛍光計測技術を用いた高分子クラウディングが与える相互作用の評価
Elucidation of the Effect of Macromolecular Crowding to Molecular Interactions using Fluorescence Fluctuation Microscopy Techniques
Fusako Gan[1], Akito Matsui[1], Johtaro Yamamoto[2], Masataka Kinjo[3] ([1]*Grad. Sch. of Life Sci., Hokkaido Univ.*, [2]*Biomed. Res. Inst., AIST*, [3]*Fac. of Adv. Life Sci., Hokkaido Univ.*)

3Pos176 Examination of backtracking engulfment mechainism in macrophages using on-chip single cell observation assay
Amane Yoshida[1], Yuya Furumoto[1], Toshiki Azuma[1], Takahiro Kitahara[2], Tomoyasu Sakaguchi[2], Masao Odaka[3], Akihiro Hattori[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

3Pos177 キャピラリー吸引法を用いた単一マクロファージの一連の貪食における余分な体積増加について
Extra volume increase of single macrophage during sequential phagocytosis occurred by using micropipette aspiration measurement assay
Toshiki Azuma[1], Yuya Furumoto[1], Amane Yoshida[1], Masao Odaka[2], Akihiro Hattori[2], Tomoyasu Sakaguchi[3], Takahiro Kitahara[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Org. Univ. Res. Initiatives, Waseda Univ.*, [3]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ*)

3Pos178 Releasing SecM translation arrest and observing resumed translation using magnetic tweezers
Zhuohao Yang, Ryo Iizuka, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

バイオイメージング／Bioimaging

3Pos179 (3SGA-5) 転写伸長を制御するメディエーターの１分子超解像イメージングによる分子局在と動態の定量解析
(3SGA-5) Molecular localization and dynamics of Mediator regulating transcription elongation using single-molecule and super-resolution microscopy
Yuma Ito[1], Shinnosuke Kunimi[1], Hidehisa Takahashi[2], **Makio Tokunaga**[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*)

3Pos180 酸性細胞環境内の超解像イメージング応用に向けた耐酸性可逆的光スイッチング緑色蛍光タンパク質の開発
Acid-tolerant Reversibly Switchable Green Fluorescent Protein for Super-resolution Imaging in Acidic Conditions
Hajime Shinoda[1,2], Kai Lu[3], Ryosuke Nakashima[3], Tetsuichi Wazawa[3], Kosuke Noguchi[2], Tomoki Matsuda[2,3], Takeharu Nagai[2,3] ([1]*CPR, Riken*, [2]*Grad. Sch. Eng., Osaka Univ.*, [3]*ISIR, Osaka Univ.*)

3Pos181 Direct observation of heterogeneous starvation response and emergence of surviving subpopulation in the clonal microbial population
Sotaro Takano[1], Miki Umetani[2], Hidenori Nakaoka[2], Yuichi Wakamoto[2,3,4], Ryo Miyazaki[1,5,6] ([1]*AIST, Bioprod. Inst.*, [2]*Grad. Sch. of Arts and Sci., Univ. of Tokyo*, [3]*Universal Biol. Inst., Univ. of Tokyo*, [4]*Res. Center for Complex Syst. Biol., Univ. of Tokyo*, [5]*AIST, CBBD-OIL*, [6]*Life and Env. Sci., Univ. of Tsukuba*)

3Pos182 ヘアレスマウスにおける皮質拡延性抑制の近赤外無侵襲測定
Noninvasive near-infrared monitoring of intrinsic optical signals caused by high K+-induced cortical spreading depression in hairless mice
Hiro Yamato[1], Takashi Jin[2], **Yasutomo Nomura**[1] ([1]*Grad. Eng. Maebashi Inst. Tech.*, [2]*RIKEN BDR*)

3Pos183 1粒子でナノスケール温度計とナノスケール熱源になる蛍光ナノダイヤモンド
Application of individual fluorescent nanodiamond as nanothermometer and nanoheater
Chongxia Zhong[1], Shingo Sotoma[1,2], Yoshie Harada[1,3], Madoka Suzuki[1] ([1]*Institute for Protein Research (IPR), Osaka University*, [2]*Japan Society for the Promotion of Science(JSPS)*, [3]*QIQB, OTRI, Osaka University*)

3Pos184 粒子フィルター MD シミュレーションによる高速 AFM データからの分子動態推定
Biomolecular dynamics inferred from high-speed AFM data via particle-filter MD simulations
Suguru Kato, Toru Niina, Sotaro Fuchigami, Shoji Takada (*Kyoto University*)

3Pos185 Elucidation of the aggregation of serum amyloid A protein and health diagnosis using a high-throughput screening system
Xuguang Lin[1], Masahiro Kuragano[1], Kenichi Watanabe[2], Kiyotaka Tokuraku[1] ([1]*Dep. of App. Sci. and Eng., Muroran Ins. of Tech.*, [2]*Obihiro Univ. of Agric. Vet. Med.*)

3Pos186 3D time-laps imaging of alpha-synuclein aggregation using quantum-dot nanoprobes
Min Nuo[1], Masahiro Kuragano[1], Q.P Taro Noguchi[2], Kiyotaka Tokuraku[1] ([1]*Dep. of App. Sci.and Eng., Muroran Ins. of Tech.*, [2]*NIT, Miyakonojo College*)

3Pos187 Three-dimensional analysis for formation process of amyloid β_{42} aggregation using quantum dots nanoprobe
Masahiro Kuragano, Kiyotaka Tokuraku (*Div. of Sust. and Env. Eng., Muroran Inst. of Tech.*)

3Pos188 高速 AFM を用いた HECT 型ユビキチンリガーゼのユビキチン化に伴う構造動態の観察
Observation of the structural dynamics associated with ubiquitination of HECT-type ubiquitin ligase using high-speed AFM
Ikumi Mruo[1], Takahiro Watanabe-Nakayama[2], Toshio Ando[2], Hiroki Konno[2] ([1]*Grad. Sch. of Nat. Sci. & Technol., Kanazawa Univ.*, [2]*WPI Nano Life Sci. Inst. (WPI-NanoLSI), Kanazawa Univ.*)

3Pos189 Kinesin transport on microtubules studied by high-speed AFM
Christian Ganser[1], Syeda Rubaiya Nasrin[2], Akira Kakugo[2,3], Ryota Iino[4], Takayuki Uchihashi[5] ([1]*ExCELLS, NINS*, [2]*Grad. Sch. Chem. Sci. Eng., Hokkaido Univ.*, [3]*Fac. Sci., Hokkaido Univ.*, [4]*IMS, NINS*, [5]*Grad. Sch. Sci., Nagoya Univ.*)

3Pos190 鎖の細胞膜提示システムの構築とその機能解析
Construction and functional analysis of the glycan display system on the cell membrane
Ayane Miura[1], Kazuya Kabayama[1,2,3], Syuto Miyake[1], Hiroki Syomura[1], Yoshiyuki Manabe[1], Toshiyuki Yamaji[4], Kentaro Hanada[4], Koichi Hukase[1,2,3] ([1]*Grad. Sch. of Sci., Osaka Univ.*, [2]*MS-CORE, PRC, Grad. Sch. of Sci., Osaka Univ.*, [3]*Inst. for Radiation Sciences, Osaka Univ.*, [4]*NIID*)

3Pos191 分岐鎖アミノ酸に対する遺伝子コード型蛍光バイオセンサー
Genetically encoded fluorescent biosensor for branched-chain amino acids
Hiromi Imamura, Tomoki Yoshida, Hitomi Nakajima, Sena Takahashi, Akira Kakizuka (*Grad. Sch. Biost., Kyoto Univ.*)

3Pos192 Odor-evoked responses in mouse whole brain as detected by BOLD-fMRI analyses with periodic stimulation and independent component analysis
Mitsuhiro Takeda, Fuyu Hayashi, Naoya Yuzuriha, Sosuke Yoshinaga, Hiroaki Terasawa (*Kumamoto University, Faculty of Life Sciences*)

3Pos193 クライオ電子線トモグラフィー法による糸状仮足中のアクチン繊維とファシンの可視化
Visualization of F-Actin with Fascin in Filopodia by electron cryo-tomography
Naoko Kajimura[1], Takuo Yasunaga[2], **Kaoru Mitsuoka**[1] ([1]*Research Center for Ultra-High Voltage EM, Osaka Univ.*, [2]*Grad. Sch. Comp. Sci. Syst. Eng., KIT*)

3Pos194 Fluorescent Ca2+ indicators for multiplexed super-resolution imaging at nanoscopic cellular domain
Kai Lu, Tomoki Matsuda, Tetsuichi Wazawa, Takeharu Nagai (*ISIR, Osaka University*)

3Pos195 (3SFA-5) Intracellular delivery of biologics using magnetically-navigated nanocarrier
Yoshihiro Sasaki, Ryosuke Mizuta, Naoya Kinoshita, Kazunari Akiyoshi (*Department of Polymer Chemistry, Graduate School of Engineering, Kyoto University*)

3Pos196 3D image construction methods for observation of cell with micro manipulation
Masaru Kojima[1], Yuma Takeuchi[1], Yasushi Mae[1], Tatsuo Arai[2,3] ([1]*Grad. Sch. Eng.Sci., Osaka Univ.*, [2]*UEC*, [3]*BIT*)

3Pos197 Distinct morphologies of integrin-targeted peptide co-assemblies in peritumoral space vs physiological ECM
William Cortes, Sona Roy, Sachie Yukawa, **Toshio Sasaki**, Wu Xia, Ye Zhang (*Ye Zhang Unit, Okinawa Institute of Science and Technology*)

3Pos198 環状柔軟多関節 DNA モチーフの自己集合
Self-assembly of a Flexible Multi-joing Ring Motif
Shiyun Liu, Ibuki Kawamata, Shin-ichiro Nomura, Satoshi Murata (*Grad. Sch. Eng., Univ. Tohoku*)

3Pos199 オン・チップマルチイメージングフローサイトメトリーでの血中の循環腫瘍細胞の同定のためのサイズ解析
Size distribution analysis of circulating tumor cell clusters in blood using on-chip multi-imaging flow cytometry
Masao Odaka[1], Akihiro Hattori[1], Kenji Yasuda[1,2] ([1]*Org. Univ. Res. Initiatives, Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*)

3Pos200 アミロイド凝集阻害物質の自動スクリーニングシステムにより見出した高活性天然抽出物の評価
Evaluation of highly active natural extracts found by an automated screening system for amyloid aggregation inhibitors
Rina Sasaki[1], Masahiro Kuragano[1], Kenji Monde[2], Koji Uwai[1], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Eng., Muroran Inst. Tech.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*)

3HSPos001 ハクセンシオマネキの日周期と親愛なる敵効果
Circadian and Dear Enemy Effect of Uca lactea
Mei Oshikawa, Ayumi Fuse, Rinka Aida (*Miyazaki Kita High School*)

3HSPos002 オカダンゴムシの移動と体重減少速度の関係
Pill-bag's relationship between movement and weight decrease rate
Takato Mera (*Miyazaki Kita High School*)

3HSPos003 アサリの潜砂率について
In what condition do the clams under the sand
Syunitiro Maruta, Sayaka Haruyama (*Miyazaki Kita High School*)

3HSPos004 レゴで昇降機の作製とその工夫
Fabrication and devising of elevator with LEGO
Kodai Hirota (*Miyazaki Kita High School*)

3HSPos005 セルロースチューブ内の結晶析出
Crystal Deposition in Cellulose Tubes
Kazuto Ozeki (*Miyazaki Kita High School*)

3HSPos006 チャコウラナメクジの重力走性と光走性の関係
Gravitational motility of slugs
Misaki Inoue, Takanori Matsuda (*Miyazaki Kita High School*)

3HSPos007 スクミリンゴ貝の習性
Investigating the Habits of apple snails
Asuka Onitsuka (*Miyazaki Kita High School*)

3HSPos008 火山灰と植物の関係性
The effect of volcanic Ash on plants
Yui Nakashima (*Miyazaki Kita High School*)

1CAP-01 急速に拡大する微生物型ロドプシンワールド
Fast-expanding microbial rhodopsin world

Keiichi Inoue (*Inst. Solid State Phys., Univ. Tokyo*)

Microbial rhodopsins are photoreceptive membrane proteins in which all-trans retinal chromophore binds. The first reported microbial rhodopsin, bacteriorhodopsin, works as light-driven outward proton pump. Subsequently, inward chloride pump and phototaxis sensors were discovered. In this century, however, the innovation of DNA sequencing technology, especially next-generation sequencers, revealed a large number of new families of microbial rhodopsins from various micro-organisms, and the functions are also diverse: ion channels, gene regulation, enzymes and so on. Furthermore, heliorhodopsin distinct from whole microbial rhodopsin family was reported in 2018. In this presentation, the current situation of the fast-expanding microbial rhodopsin world will be discussed.

1CAP-02 生体光物理の挑戦：静的な孤立系から組織化された動的な集合体の理解へ
Next challenge in biological photophysics: From static/isolated protein to dynamic/assembled protein network

Toru Kondo (*Tohoku University*)

Biological systems are driven by multiple proteins organizing a functional network. To understand the regulation mechanism, each protein is biochemically isolated and then examined. In particular, the photoreaction in chromoprotein has been interpreted within the context of the simplified situation. However, proteins are not independent but incorporated into the system. Additionally, while the x-ray protein crystallography enables a detailed discussion based on the protein structure, at physiological conditions, the protein conformation can fluctuate thermally or change as a result of the photoreaction. Thus, it is an essential question whether the overall function of the biological system is just a linear sum of each function of “static” and “isolated” proteins or not.

1CAP-03 CRISPR-Cas9 の立体構造と機能改変
Structure and Engineering of CRISPR-Cas9

Hiroshi Nishimasu (*The University of Tokyo*)

The CRISPR RNA-guided DNA endonuclease Cas9 cleaves a double-stranded DNA complementary to the RNA guide, and has been harnessed for genome-editing applications. Cas9 requires a protospacer adjacent motif (PAM) for the target recognition. We determined the crystal structures of the Cas9-guide RNA-target DNA complex, and provided mechanistic insights into the RNA-guided DNA cleavage by Cas9. Furthermore, we used high-speed atomic force microscopy to visualize the dynamics of CRISPR-Cas9, such as the fluctuation of the HNH nuclease domain upon DNA binding. More recently, we rationally engineered a Cas9 variant (SpCas9-NG) that can recognize a relaxed NG, rather than NGG, as the PAM, and thus expanded target ranges in Cas9-mediated genome engineering.

1CAP-04 生物分子モーターとその集合体の再デザイン
Re-design of biomolecular motors and their systems

Ken'ya Furuta (*NICT*)

During the past decade, knowledge about the mechanisms of protein-based biomolecular motors has markedly increased. However, it is presently not possible to design a new biomolecular motor, much less its collective behavior that leads to various functions within the cell. Recent achievements in the biomolecular motor field include re-engineering of individual motors, construction of multi-motor systems, and generation of large-scale complex behavior. Here we focus on the former two levels of complexity. We propose here a strategy where the collective behavior can be repeatedly tested upon modifying individual motors, which may provide important clues about how biomolecular motors and their systems are designed.

1CAP-05 相分離研究の最近の動向
Current trend of phase separation

Tomoshi Kameda, Yoichi Kurumida, Yusuke Nakamichi, Keisuke Ikeda, Ryo Kitahara, Kiyoto Kamagata (*AIRC, AIST*)

Recently, many researchers have started studying phase separation (PS) of protein and nucleic acid. PS includes aggregation, fibrillization(solid), gelation and liquid-liquid PS (LLPS). These phenomena are open and interesting problems in term of physics. In a biological point of view, PS is also interesting topic because it plays an important role in a variety of cellular processes, such as formation of classical membrane-less organelles, signaling complexes, stress granules and numerous supramolecular assemblies. And, PS is known to be relevant to human diseases, especially nervous disease. In this talk, I will review current trends and show our study about PS.

2CAA-01 バイオメディカル、細胞生物学、生理学分野におけるナノ温度計測の近況
Current nanothermometry in cell biology and physiology

Madoka Suzuki (*Institute for Protein Research, Osaka University*)

Nanothermometry is a way to determine the temperature or its changes in a "small" volume such as a cell. Some methods of nanothermometry rely on microfabrication technologies. Others use luminescent dyes or nanomaterials as temperature probes that require optical microscopes to measure signals from them. In this presentation, I will introduce current progress of these methods and their applications to biomedical studies, cell biology and physiology by showing examples including those from our group.

2CAA-02 細胞の中ではたらく物理的な力
Physical forces in the cell

Hirokazu Tanimoto (*Yokohama City University*)

We will discuss recent studies of intracellular mechanics with an emphasis on direct force measurements.

2CAA-03 The mechanics of nature behind the multicellular tissue structure

Kaoru Sugimura (*Institute for Integrated Cell-Material Sciences (WPI-iCeMS), Kyoto Univ.*)

Let’s consider a bridge made of blocks. A mechanic or engineer carefully calculates forces acting between individual blocks to construct a bridge that does not fall down. For instance, in a half circle bridge, the block on the top of a half circle, which is called a corner stone, is known to be critical to keep its shape. Animals and plants have cells instead of blocks. During human development, forces acting between trillions of cells shape our body. Thus, to solve a mystery of diverse forms in nature, we need to understand mechanical interaction between cells. In this talk, I will review what we know about the mechanics of nature behind the multicellular tissue structure and discuss future directions and challenges for the field.

2CAA-04 細胞外体液動態を介した分泌タンパク質の新たな制御機構
Novel extracellular fluid mechanism for regulation of secreted proteins in Xenopus laevis

Hidehiko Inomata (*RIKEN BDR*)

Secreted proteins, such as morphogens, provide positional information for embryonic cells in a concentration-dependent manner, hence their distribution and activity are strictly regulated in the embryo. Secreted proteins synthesized from source cells diffuse into the extracellular space and are degraded by mechanisms such as protease-mediated degradation or receptor-mediated endocytosis. In addition, secreted proteins are regulated by specific inhibitors and transport mechanisms, such as transcytosis and cytonemes. However, it remains unknown how extracellular fluid, which contains secreted proteins, affects their distribution and activity. Here, we introduce a novel role of extracellular fluid dynamics in secreted protein-mediated tissue patterning in Xenopus laevis.

2CAA-05 Recent Advances in Origins of Life Research by Biophysicists

Tony Z. Jia (*Earth-Life Science Institute, Tokyo Institute of Technology and Blue Marble Space Institute of Science*)

Although most biophysics research is focused on extant biology, at the core of modern biology is the question of how the earliest life on (or off) Earth emerged. Recent technological and methodological advances developed by biophysicists have allowed researchers to gain new knowledge related to the origins of life (OoL). Here, we highlight some of the significant OoL advances contributed by biophysists with respect to the synthesis and assembly of biological (or pre-biological) components on early Earth, co-assembly of primitive compartment, and evolution of early genetic systems. We hope to provide inspiration to other biophysicists to consider how their interdisciplinary work or techniques can contribute to the ever-growing field of OoL research.

2CAP-01 高分子混雑と細胞サイズ閉じ込めの協奏による特異な相挙動
Unique phase behavior in cell size space: Synergistic effect of molecular crowding and confinement

Miho Yanagisawa (*Graduate School of Arts and Sciences, The University of Tokyo*)

In living cells, highly concentrated molecules are confined in biomembranes. Through studies using artificial cells, the unique phase behaviors of high concentration molecules confined in cell size space have been elucidated (Yanagisawa, et al., 2014, Int. Rev. Cell. Mol. Biol.; Watanabe & Yanagisawa, 2018, PCCP.; Sakai, et. al., 2018 ACS Cent. Sci.). In this talk, I will explain these behaviors from the view of synergetic effect between macromolecular crowding (excluded volume effect, steric repulsion, etc.) and cell size confinement (small volume and membrane effects). I will also introduce the contribution from soft matter physics for elucidation of the mechanism of various phase transitions in cells such as liquid-liquid phase separation.

2CAP-02 合成生物学は生物実験をどう変えるか　―生物物理学の視点から―
“Dream” experiment in living cells? Synthetic biology may help in near future!

Hideki Nakamura (*The Johns Hopkins University*)

Synthetic biology aims at understanding life by synthetically recapitulating or manipulating it. Recent expansion of methodologies in the field would be beneficial not only for experimental biologists but also for theorists, providing more opportunities for models and concepts to be tested experimentally. In the talk, I will first summarize recent innovations in synthetic biology. I then focus on techniques that manipulate fast biological processes, introducing two tools I developed. These tools, iPOLYMER and ActuAtor, manipulate biophysical processes in living cells, suggesting the promise of synthetic biology in validating models proposed by theories or by in vitro studies. Open challenges and future perspectives will also be discussed.

2CAP-03 神経細胞軸索輸送の分子モーターカウンティング
Molecular motor counting for neuronal cargo transport

Kumiko Hayashi[1, 2] ([1]*Sch. Eng., Tohoku Univ.*, [2]*PRESTO, JST*)

Neuronal morphology necessitates particularly fast cargo vesicle transport for efficient communication between the cell body and distal processes. Molecular motors such as kinesin superfamily proteins and cytoplasmic dynein haul cargo vesicles anterogradely toward the terminal and retrogradely toward the cell body. We clarified that several molecular motors carried a single cargo vesicle cooperatively to keep the stability and efficiency of the transport based on a fluorescence image analysis using non-equilibrium statistical mechanics. This non-invasive molecular motor counting method enables to clarify the molecular basis of neuronal diseases.

2CAP-04 多細胞系の非平衡物理モデルと実験
Physical properties of homeostatic and active tissues

Kyogo Kawaguchi (*RIKEN CPR*)

Turnover and migration of cells are the two main factors that drive multicellular tissues out of equilibrium. In adult tissue homeostasis, a typical system undergoing turnover, a certain model of stochastic interacting particles has been shown to fit with the live imaging and clonal tracing data. On the other hand, multiple examples of migratory cells at high density have been reported to exhibit macroscopic features of active matter. Here we will discuss why some properties of theoretical models are robust enough to be observed in these experiments, and show how these properties can be used to learn about the details of biology such as the nature of cell-to-cell interactions.

2CAP-05 細胞建築学：物理学と遺伝学の一交差点
Architectonics of the cell, as a crossroad of physics and genetics

Akatsuki Kimura[1, 2] ([1]*Cell Arch Lab, National Institute of Genetics*, [2]*Dept Genetics, SOKENDAI*)

Physics is becoming more important in modern cell biology. To understand how cells –which are organized architectures– are constructed, I believe a true collaboration between physics and genetics is inevitable. Measurements and calculations of mechanical forces should be combined with efforts in genetics to characterize gene products. The construction of the cell from its molecular components is considered as a self-organization process, and thus concepts from non-equilibrium physics are expected to take us beyond our view of the cell as a ‘molecular machine.’ Having the abovementioned belief in mind, my research group has been working on constructing mechanical models for cell-scale behaviors such as nuclear centration, cytoplasmic streaming, and cell division.

2CAP-06 神経ダイナミクスの埋め込みと予測
Embedding and Predicting Neural Dynamics

Taro Toyoizumi (*RIKEN Center for Brain Science*)

Brain-wide interactions generating complex neural dynamics are considered crucial for emergent cognitive functions. However, the irreducible nature of nonlinear and high-dimensional dynamical interactions challenges conventional reductionist approaches. We introduce a model-free method, based on embedding theorems in nonlinear state-space reconstruction, that permits a simultaneous characterization of directed network interactions and complexity in local dynamics. First, we apply this method in large-scale electrophysiological recordings from awake and anesthetized monkeys. The method reveals a hierarchy of cortical areas specifically in awake conditions, where dynamical complexity increases along with cross-area information flow. Second, in cultured neural networks, we detect the "early warnings" of a synchronous population burst based on its local dynamics, demonstrating that seemingly stochastic bursting events can be accurately predicted by near deterministic dynamics of single neurons. This prediction based on local dynamics can even outperform the predictions based on population activity. We explain this apparently counterintuitive property by the network structures realizing in the non-bursting period, which is supported by a manipulative experiment and analyses. These findings demonstrate the advantages of the embedding method in deciphering large-scale and heterogeneous neural systems.

3CAA-01 X線自由電子レーザーによるタンパク質分子動画撮影
Molecular movies of proteins at work by X-ray free electron lasers

Eriko Nango[1,2], So Iwata[1,2] ([1]*Graduate School of Medicine Kyoto University*, [2]*RIKEN SPring-8 Center*)

Serial femtosecond crystallography (SFX) is an emerging technique for determining a protein crystal structure using X-ray free-electron lasers (XFEL) and allows data acquisition at room temperature as intense femtosecond X-ray pulses from XFEL afford diffraction patterns from microcrystals before the onset of radiation damage. Furthermore, this technique enables to capture structural changes in proteins at high spatial and temporal resolution by the combination of a reaction initiator such as light excitation. Recently, we revealed structural changes in bacteriorhodopsin during proton transfer from femtoseconds to milliseconds using time-resolved SFX. We will present the latest results from our time-resolved SFX studies and future prospects.

3CAA-02 クライオ電子顕微鏡法による生物試料の構造解析
Structural Analysis of Biological Samples by Cryogenic Electron Microscopy

Kaoru Mitsuoka (*Research Center for Ultra-High Voltage Electron Microscopy*)

Possible resolutions by structural analysis of biological samples using cryogenic electron microscopes (cryo-EM) has improved drastically by recent technical improvements. Now single-particle cryo-EM is routinely used to determine atomic models of biomolecules and their complexes. In addition, their structures in cells can be visualized by cryo-electron tomography (cryo-ET). Thus, we show two applications of cryo-EM, one is V-ATPase by single-particle cryo-EM and the other is a complex of facin and actin filaments in cells by cryo-ET. By single-particle cryo-EM, we could determine several structures related to the function of V-ATPase. By cryo-ET, we could fit atomic models of facin and actin to the actin-filament bundles in neuron-model cells.

3CAA-03 生細胞の超解像度機能イメージング
Super resolution live cell functional imaging

Yasufumi Takahashi (*Kanazawa university, NanoLSI*)

Scanning ion conductance microscopy (SICM) is a unique and promising technique for super resolution live cell topographical imaging under physiological conditions. SICM uses a nanopipet as a probe to detect the ionic current between an electrode inside the pipet and an electrode located in a bath and provides noncontact topography image of live cell. The pipet–sample distance is regulated by the ionic currents used as feedback signals. SICM is effective tool for linking the topography and ion channel distribution by navigating the nanopipette on cell surface using topography information and perform region specific patch clamp recording. Nanopipette is also useful tool for chemical deliver and collection at region specific cell function.

3CAA-04 機械学習が駆動する次世代ハイコンテンツ解析で何を「見る」か
Machine learning-driven high content analysis "Beyond seeing is believing

Sadao Ota (*Research Center for Advanced Science and Technology*)

In this talk, I'd like to discuss not only our recent works related to an image-free imaging cell sorter, named ghost cytometry, but also what we are prospecting beyond conventional imaging in the era of machine learning. "Seeing is believing" has been a popular phrase in imaging community for a very long time, and will be important in the future as well. In some (becoming many) cases for a variety of imaging technologies, however, use of powerful machine learning technologies may expand our "ability of seeing" by data-driven prediction. As just one example, our ghost cytometry showed how to use multi-modal measurements and machine learning methods for a specific purpose of "high speed cell classification based on image information".

3CAA-05 大脳皮質から脳深部までの包括的な脳活動計測が可能な完全埋め込み型フレキシブル・ストレッチャブ集積回路システム
Brain-implanted flexible and stretchable integrated circuit system for comprehensively monitoring brain activities from cerebral cortex to deep brain regions

Tsuyoshi Sekitani (*The Institute of Scientific and Industrial Research, Osaka University*)

To totally understand brain-function networks, monitoring of brain signals by placing brain-signal sensors at various positions from the cerebral cortex to deep brain regions using advanced technology is important. However, the brains of most primates with advanced brain activities are small and are easily compressed by these sensors. Therefore, the development of a thin, flexible, and lightweight sensor system is indispensable. In this study, we have developed a brain-implanted multichannel system for monitoring brain activities using (1) thin, flexible, and stretchable integrated circuits and sensors and (2) a highly conducting and flexible material with high stretchability like rubber and an organic integrated circuit technology using this material.

1SBA-1 Chromatin dynamics and transcription

Kazuhiro Maeshima, Kayo Hibino, Ryosuke Nagashima (*Genome Dynamics Laboratory, National Institute of Genetics*)

Chromatin organization and dynamics play a critical role in gene transcription. However, how they interplay remains unclear. Using the single nucleosome imaging technique, we investigated genome-wide chromatin behavior under various transcriptional conditions in living human cells. While transcription by RNA polymerase II (RNAPII) is generally thought to need more open and dynamic chromatin, surprisingly, we found that active RNAPII globally constrains chromatin movements. RNAPII inhibition or RNAPII rapid depletion released the chromatin constraints and increased chromatin dynamics. Our results demonstrated that chromatin is globally stabilized by loose connections through active RNAPII.

1SBA-2 真核生物における転写メディエーター複合体の構造機能解析
Structural and functional basis of the Mediator complex in the eukaryotic transcriptional system

Kayo Nozawa (*The University of Tokyo, Institute for Quantitative Biosciences*)

Mediator is a large co-activator of eukaryotic transcription that directly connects activator which is bound to regulatory DNA elements with RNA polymerase II. The Mediator system is required for the majority of protein coding gene expressions. In yeast, Mediator comprises 25 subunits with a total mass of ~1.4 MDa and is organized into four modules, called head, middle, tail and kinase. The head and middle modules form the essential core of Mediator, whereas the tail and kinase modules play regulatory roles. Various structural studies including our work revealed almost entire structure of Mediator, leaving tail module and kinase module as last unknown part. In this talk, I would like to summarize recent findings within Mediator that are important for gene regulation.

1SBA-3 RNA ポリメラーゼ II によるヌクレオソーム転写のメカニズム
Structural basis of nucleosome transcription by RNA polymerase II

Shun-ichi Sekine (*RIKEN BDR*)

In eukaryotic cells, genomic DNA forms chromatin, composed of multiple nucleosomes. The nucleosomes are inherent roadblocks for gene transcription by RNA polymerase II (RNAP II), a huge protein complex responsible for mRNA transcription. The mechanism by which RNAP II transcribes the nucleosomal DNA had been a long-standing question in biology. We analyzed the structures of the nucleosome-transcribing RNAP II with and without transcription elongation factors by cryo-electron microscopy (1,2). The structures revealed how RNAP II progresses through a nucleosome, as well as how elongation factors facilitate the efficient RNAP II progression.

1. Kujirai et al. (2018). doi: 10.1126/science.aau9904
2. Ehara et al. (2019). doi: 10.1126/science.aav8912

1SBA-4 クライオ電顕と MD 計算による RNA ポリメラーゼ II 伸長状態複合体の動態解析
Dynamic structures of the RNA polymerase II elongation complex by cryo-EM and MD approaches

Takaharu Mori[1], Haruhiko Ehara[2], Shun-ichi Sekine[2], Yuji Sugita[1,3,4] ([1]*RIKEN Theor. Mol. Sci. Lab.*, [2]*RIKEN BDR (Yokohama)*, [3]*RIKEN BDR (Kobe)*, [4]*RIKEN R-CCS*)

RNA polymerase II has a central rule in the first step of the central dogma. The transcription is mainly composed of three steps: initiation, elongation, and termination. Recently, the elongation complex structure was determined by cryo-EM. However, several parts of the structures were still unrevealed due to low local resolution caused by large fluctuations. In this study, we modeled such parts based on the molecular dynamics based flexible fitting. We analyzed dynamics of DNA in the pretense and absence of the transcription factors.

1SBA-5 Allostery of Nucleosomal DNA for Transcription Factor Binding

Cheng Tan, Shoji Takada (*Kyoto University*)

Recent experiments have revealed that some transcription factors (TFs) can bind to and affect the stability of nucleosomes. Here we use coarse-grained molecular dynamics simulations to study the nucleosome-binding of two TFs, Sox2 and Oct4. Our simulations show that Sox2 energetically selects DNA motifs exquisitely phasing outward, or entropically binds to the dyad, where more possible rotational positions exist. We also find that the two domains of Oct4 can bind on the acidic patch or the two parallel gyres of nucleosomal DNA. By simulating multiple TFs binding on nucleosome, we find a DNA allosteric mechanism, by which the binding of one Sox2 can change the rotational position of DNA and affect the binding probability of the second approaching Sox2 or Oct4.

1SBA-6 (1Pos082) 大腸菌非六量体型 DNA ヘリカーゼ UvrD C 末端欠損変異体の 1 分子イメージング (1Pos082) Single-molecule imaging of a non-hexameric *Escherichia coli* helicase UvrD mutant lacking C-terminal residues

Hiroaki Yokota (*Biophotonics Lab., Grad. Sch. Creation New Photon. Indust.*)

Escherichia coli UvrD protein is a non-hexameric superfamily I DNA helicase which plays a crucial role in nucleotide excision repair and methyl-directed mismatch repair. We revealed by single-molecule fluorescence imaging that wildtype UvrD unwinds DNA in the form of an oligomer (dimer or trimer) (*Biophys. J.* 2013). Although it has been reported that C-terminus residues are crucial to dimerization and unwinding activity of UvrD, their role is poorly understood. Thus, in this study, single-molecule direct visualization was performed for a UvrD mutant lacking C-terminal residues. Contrary to the monomeric model proposed by biochemical, genetic and X-ray crystallography studies using the mutant, this study showed that the mutant also unwound DNA in the form of an oligomer.

1SCA-1 DNA origami lattices self-assembled on lipid bilayer membranes

Yuki Suzuki (*FRIS, Tohoku Univ.*)

Molecular self-assembly has attracted great attention as a method to design and construct novel supramolecular architectures. In this study, DNA origami components are self-assembled into two-dimensional lattices on an artificial lipid bilayer membrane. The adsorption of DNA origami components onto the fluidic lipid bilayer allows their surface diffusion, facilitating interaction among the components to assemble into micrometer-sized lattices in their lateral dimensions. The lattices interacting with lipid bilayers will serve as a versatile platform for a diverse range of applications, including a periodic arrangement of protein molecules or nanoparticles, scaffolds for nanodevices, and mimics of membrane-cytoskeleton networks.

1SCA-2 (1Pos074) 脂質分子の混み合い効果による膜貫通タンパク質結晶化の検討 (1Pos074) Crystallization of transmembrane protein driven by molecular crowding effect of lipids: Theoretical estimation by using a simple model

Keiju Suda[1], Ayumi Suematsu[2], Ryo Akiyama[1] ([1]*Kyushu University, Sci.*, [2]*Kyushu Sangyo University, Science and Engineering*)

We studied driving force of transmembrane protein crystallization by using a two-dimensional binary hard disk system, namely a mixture of lipids and proteins. Wild type bacteriorhodopsin (bR) trimers crystallize in a lipid bilayer. On the other hand, some mutant bRs do not construct trimers, and the critical concentration(CC) of crystallization is 10.2 times higher than wild type bRs. The free energy curves are obtained based on the free volume theory and the scaled particle theory and phase diagrams are drawn. The phase diagrams for trimer- and monomer- lipid systems are compared and the ratio between CCs (CCR) are obtained. Calculated CCR agrees with the experimental one. This result suggests that the depletion force plays an important role in the crystallization.

1SCA-3 生理学的等温条件下における細胞サイズリポソーム内での特定配列を持つ DNA 分子の増幅
Amplification of specific DNA molecules inside giant unilamellar vesicles at isothermal and physiological temperature

Yusuke Sato[1], Ken Komiya[1], Ibuki Kawamata[2], Satochi Murata[2], Shin-ichiro M. Nomura[2] ([1]*Sch. Comput. Tokyo Tech*, [2]*Grad. Sch. Eng., Tohoku Univ.*)

The amplification of DNA in environments surrounded by lipid membranes provides fundamental techniques in the creation of artificial biological systems. Here, we report amplification of specific DNA molecules in giant unilamellar vesicles (GUVs) at an isothermal temperature of 37°C. We designed the amplification circuit that can produce and amplify “output” DNAs from only a small quantity of signal DNA. Using this circuit, over 5000-fold amplification of output DNAs was achieved inside GUVs. Furthermore, we demonstrated initiation of the amplification can be triggered by photo-stimulation. We believe that our results will become the basis for the creation of novel molecular systems that regulate the functions of the system using the amplified DNA signals.

1SCA-4 (1Pos078) cDNA ディスプレイとセルソーターの利用による新規リポソームポア形成ペプチドの創製
(1Pos078) Novel pore-forming peptides assembling in liposome membranes selected by combining cDNA display method with cell sorter system

Naoto Nemoto[1], Toshiki Miyajima[1], Takeru Yoshinobu[1], Yusuke Sekiya[2], Ryuji Kawano[2] ([1]*Grad. Sci. Eng., Saitama Univ.*, [2]*Dept. Biotech. Life. Sci., Tokyo Univ. Agr. Tech*)

Previously, we have selected some liposome-binding peptides by in vitro selection using cDNA display. cDNA display method is a genotype-phenotype linking method with a cell-free translation system by fusing a cDNA with its coding polypeptide via a puromycin covalently. Owing to its stability and robustness, cell-based selections can be performed with this technology just like phage display. In this study, we performed an in vitro selection of pore-forming peptides which act functionally like antimicrobial peptides in liposome membranes, by using cDNA display with a Fluorescence Activated Cell Sorting system. Interestingly, the selected peptides form channels with ranging from 2 to 5 nm diameter in the liposome membrane by analyzing high-throughput lipid bilayer system.

1SCA-5 (1Pos287) 光からエネルギーを合成しタンパク質合成をする人工光合成細胞の構築
(1Pos287) Artificial photosynthetic cell producing energy for protein synthesis

Samuel Berhanu[2], Takuya Ueda[3], **Yutetsu Kuruma**[1] ([1]*JAMSTEC*, [2]*ELSI, Titech*, [3]*Grad. Sch. of Front. Sci., Univ. of Tokyo*)

Construction of an artificial cell widens our understanding of living organisms, but the mechanism to synthesize its own constituents by self-sufficient energy has not been developed so far. Here, we constructed an artificial cell that produces ATP by light and synthesizes its own constituent proteins. The artificial cell contains a photoreactive organelle which artificially consists of ATP synthase and bacteriorhodopsin. The photo-synthesized ATP eventually drove the synthesis of the components of the organelle, resulted in enhancement of the organelle activity through the positive feedback of the products. Our artificial photosynthetic cell system paves the way to construct an energetically independent artificial cell.

1SCA-6 ベシクル型細胞モデルにおけるこみあい効果
A study of crowding effect in a cell model using a statistical mechanics approach

Yuno Natsume[1,2] ([1]*Fac. Sci., Japan Women's Univ.*, [2]*ExCELLS*)

A living cell contains high concentrations of macromolecules. Under such crowding conditions, inner molecules in a cell increase their available volumes by folding or aggregating, which, in turn, affect cellular functions. To investigate the entropic effect in a cell, giant vesicles have been widely used as an artificial-cell-model platform because of their cell-like membrane structure and size. Here, we show a giant vesicle that encapsulates micrometer-sized particles, which mimic cellular macromolecules. We would like to emphasize that the giant vesicle exhibits spontaneous localization of its inner particles and membrane deformation. I will discuss these remarkable experimental phenomena mainly from the viewpoint of statistical mechanics.

1SCA-7 Construction of cell-containing synthetic vesicles for bottom-up synthetic biology

Masamune Morita (*Biomed. Res. Inst. (BMRI), AIST*)

There has been a growing interest among “bottom-up synthetic biology” to design and develop vesicle-based artificial cells. What type of artificial cells can be designed in the future? From a researcher's perspective, various types of artificial cells can be proposed based on each unique idea. This presentation describes the current state of an artificial cell-living cell hybrid system project; a new variety of artificial cells that can be developed by encapsulating living cell inside a synthetic vesicle. The original idea was inspired by the theory of “Symbiogenesis” that is considered to be the origin of eukaryotic cells. This project is an important milestone in the development of artificial cells because it demonstrated proliferation of cells inside vesicles.

1SCA-8 計算折り紙による 3 次元細胞立体構造
3D Cell Structure Optimized by Computational Origami

Kaori Kuribayashi-Shigetomi (*Hokkaido University*)

The purpose of this research is to establish a technique to efficiently fold the intended 3D structures using cells. Specifically, we utilize the method of computational origami to model folding characteristics of the cells, to make an algorithm for the selection of the optimal development of the 3D shape, and to efficiently fold it with the cells. There are various three-dimensional structures and organs in the living body, and the shape plays an important role in its function. Here, we examined the difference in differentiation when cells were cultured in 2D plane and 3D structure. We also examined differences in cell shape and cytoskeleton due to differences in developments.

1SDA-1 Structural transition of nucleosome during RNA polymerase II transcription revealed by cryo-EM

Tomoya Kujirai[1,2], Haruhiko Ehara[2], Mikako Shirouzu[2], Shun-ichi Sekine[2], Hitoshi Kurumizaka[1,2] ([1]*IQB, Univ. of Tokyo*, [2]*RIKEN BDR*)

Genomic DNA forms chromatin, in which the nucleosomes is the basic unit. In chromatin, transcription of genes occurs through nucleosomes by RNA polymerase II. To reveal the mechanism how RNA polymerase II transcribes nucleosomal DNA, we established in vitro transcription system on a nucleosome template and determined the 3D structures of transcribing RNA polymerase II-nucleosome complexes using cryo-EM single particle analysis. These structures represent the snapshots of transcription reaction intermediates and reveal the structural transition of the nucleosome during RNA polymerase II passage. We will discuss potential of cryo-EM analysis for understanding transcription process on the nucleosome.

1SDA-2 タンパク質の柔軟な構造を高速原子間力顕微鏡で可視化する
Visualizing flexibility in protein structures by high-speed atomic force microscopy

Mikihiro Shibata[1,2] ([1]*WPI-NanoLSI, Kanazawa Univ.*, [2]*InFiniti, Kanazawa Univ.*)

Structural biology revealed a three-dimensional structure of proteins at the atomic level, and provided insights into a molecular mechanism of proteins. In addition to this, direct imaging of protein flexibility could enhance our understanding of how proteins function. High-speed atomic force microscopy (HS-AFM) is a capable technique to directly visualize flexible sites of single-molecules under near physiological conditions at ~ 100 millisecond time scale. In this symposium, we will present our latest HS-AFM studies of proteins including, CRISPR-Cas9 [M. Shibata *et al.* **Nat. Commun.**, 8, 1430 (2017)], hepatocyte growth factor [K. Sakai *et al.* **Nat. Chem. Biol.**, 15, 598-606 (2019)], and calcium/calmodulin-dependent protein kinase II.

1SDA-3 クライオ電子顕微鏡解析によって明らかになったミトコンドリア膜透過装置の構造と機能
Near-atomic resolution structure of the mitochondrial protein import gate

Yuhei Araiso[1], Akihisa Tsutsumi[2], Kenichiro Imai[3], Haruka Sakaue[4], Takuya Shiota[5], Kaori Yunoki[4], Junko Suzuki[4], Shin Kawano[4], Masahide Kikkawa[2], Toshiya Endo[4] ([1]*Grad. Sch. of Med. Sci., Kanazawa Univ.*, [2]*Grad. Sch. of Med., Univ. of Tokyo*, [3]*AIST*, [4]*Fac. of Life Sci., Kyoto Sangyo Univ.*, [5]*OPTT, Univ. of Miyazaki*)

The TOM complex is the main entry gate for most mitochondrial proteins. Here we report the cryo-EM structure of the yeast TOM complex, revealing the detailed architecture of an α/β translocator. The TOM complex forms a dimer and each β-barrel channel of Tom40 is tethered by two molecules of α-helical transmembrane segment of Tom22 and one phospholipid molecule in between. Combining with biochemical functional analyses, we revealed that each Tom40 channel possesses two precursor exit sites: in the middle of the dimer, a *trans* presequence-binding site is formed by Tom22/Tom40/Tom7 for presequence preproteins and at the dimer periphery, the N-extension of Tom40 and Tom5 forms an exit site for presequence-less preproteins.

1SDA-4 Microsystem for single molecule analysis of membrane proteins

Rikiya Watanabe (*CPR, RIKEN*)

Micro-chamber arrays enable highly sensitive and quantitative bioassays at the single-molecule level. Accordingly, they are widely used for ultra-sensitive biomedical applications, e.g., digital PCR and digital ELISA; however, the versatility of micro-chambers is generally limited to reactions in aqueous solutions. To address this issue, we recently developed novel microsystems using arrayed micro-sized chambers sealed with lipid bilayers for single molecule analysis of membrane proteins. In this symposium, I would like to introduce recent progress on the single molecule analysis of membrane transport proteins using the microsystem, and discuss the future prospects for its use in analytical and pharmacological applications.

1SDA-5 Cryo-EM structures of photosystem II-antenna supercomplexes

Fusamichi Akita[1,2], Ryo Nagao[1], Koji Kato[1], Naoyuki Miyazaki[3], Jian-Ren Shen[1] ([1]*RIIS, Okayama Univ.*, [2]*PRESTO, JST*, [3]*TARA, Univ. Tsukuba*)

Photosynthetic apparatuses consist of huge membrane protein complexes functioning in light-harvesting and electron transfer reactions. These complexes are hard to prepare and crystallize. The improvement in single particle cryoEM analysis technique has brought significant changes in structural biology, since it can solve the structure with a small amount of sample without crystallization. Among the photosynthetic apparatuses, Photosystem II (PSII) catalyzes the light-induced water-splitting reaction, and the PSII core is associated with antennae proteins, fucoxanthin chlorophyll a/c-binding proteins (FCPs) in diatoms and light harvest complexes (LHCs) in green plants. In this meeting, we will report some structures of the PSII-antenna complexes from different organisms.

1SDA-6 光合成型複合体 I がフェレドキシン依存性を示す構造基盤
Structural Basis for the Ferredoxin-dependency of Photosynthetic Complex I

Genji Kurisu[1], Hideaki Tanaka[1], Jan M. Schuller[2], Tsuyoshi Konuma[3], Takahisa Ikegami[3], Marc M. Nowaczyk[4] ([1]*Inst. Prot. Res., Osaka Univ.*, [2]*Max Planck Institute of Biochemistry*, [3]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*, [4]*Ruhr University Bochum*)

In cyanobacteria and chloroplasts, the oxygenic photosynthetic complex I (NDH-1L) plays a crucial role in cyclic electron flow around Photosystem I. We have determined a 3.3 A cryo-EM structure of NDH-1L from T. elongatus. Flash-induced absorption and electron paramagnetic resonance spectroscopy of isolated NDH-1L demonstrate that the complex functions as a Ferredoxin (Fd):Plastoquinone oxidoreductase, instead of using intermediates such as NADPH. To assess the sites and mode of interaction between Fd and NDH-1L, we performed NMR chemical shift perturbation experiments using 15N-labelled Fd or NdhS subunit of NDH-1L. Our integrated structural work reveals that structural adaptations of NDH-1L that facilitate binding and electron transfer from Fd.

1SEA-1 1 分子超解像局在顕微鏡法による転写装置とクロマチン構造の相互作用解析
A single-molecule localization approach to quantify the interaction between transcriptional machinery and chromatin structure

Yuma Ito, Makio Tokunaga (*Sch. Life Sci. Tech., Tokyo Tech*)

Chromatin structure has been shown to play key roles in transcriptional regulation. Although various histone variants and post-transcriptional modifications are involved in the regulation of gene expression, visualization of their functions on chromatin dynamics in living cells remains challenging. We performed single-molecule and super-resolution imaging of RNA polymerase II (Pol II) and histone variants in living cells. Localization and trajectory analysis revealed that the chromatin-bound Pol II was localized adjacent to the dense chromatin nanostructures. The spatial correlation of Pol II with chromatin was different among histone variants, suggesting a highly organized chromatin configuration that may recruit and regulate the transcriptional machinery.

1SEA-2 単一ヌクレオソームイメージングで迫る分裂期染色体の構築原理
Single nucleosome imaging reveals the physical aspect of the mitotic chromosome condensation

Kayo Hibino[1,2], Kazuhiro Maeshima[1,2], Yuji Sakai[3] ([1]*National Institute of Genetics*, [2]*SOKENDAI*, [3]*Grad. Sch. Med., Univ. Tokyo*)

Mitotic chromosomes are essential structures for the faithful transmission of copied genome information during mitosis and the subsequent various genome functions during interphase. The chromosome condensation requires some large proteins including condensin and topoisomerase IIα. However, how a long DNA is assembled into mitotic chromosomes remains unclear. Here, we measured local nucleosome dynamics during the condensation process by single nucleosome imaging in living human cells. We then investigated possible roles of condensin in the process in terms of local nucleosome behavior. Combining with simulation studies, we will discuss the physical aspect of the mitotic chromosome condensation.

1SEA-3 (1Pos095) オリゴペプチドのアミノ酸配列は DNA compaction と転写活性に著しい違いを引き起こす
(1Pos095) Marked Difference in DNA Compaction and Transcription is Caused by Amino Acid Sequence of Oligopeptide

Tatsuo Akitaya[1], Hiroyuki Hiramatsu[2], Hideaki Yamaguchi[3], Koji Kubo[4], Shizuaki Murata[4], Toshio Kanbe[5], Norio Hazemoto[6], Kenichi Yoshikawa[7], Anatoly Zinchenko[4] ([1]*Asahikawa Med. Univ.*, [2]*Fac. Pharm., Meijo Univ.*, [3]*Fac. Agr. Sci., Meijo Univ.*, [4]*Grad. Sch. Env. Std., Nagoya Univ.*, [5]*Grad. Sch. Med., Nagoya Univ.*, [6]*Grad. Sch. Pharm. Sci., Nggoya City Univ.*, [7]*Fac. Bio. Med. Sci., Doshisah Univ.*)

Compaction of T4 phage DNA (166 kbp) by short oligopeptide octamers composed of two types of amino acids, four cationic lysine (K), and four polar nonionic serine (S) having different sequence order was studied by single-molecule fluorescent microscopy. Efficient DNA compaction by oligopeptide octamers depends on the geometrical match between phosphate groups of DNA and cationic amines. The mechanism of DNA compaction changes from a discrete all-or-nothing coil-globule transition induced by a less efficient (K4S4) octamer to a continuous compaction transition induced by a (KS)4 octamer with a stronger DNA-binding character. Marked difference in the morphology of the folded DNA and the transcription activity were induced corresponding to the DNA compaction mechanism.

1SEA-4 クロマチンループを形成しないヌクレオソーム排他 DNA 配列によるインスレーター活性
Insulator Activities of Nucleosome-Excluding DNA Sequences Without Chromatin Loop Formations

Akinori Awazu[1], Yuki Matsushima[2], Naoaki Sakamoto[1] ([1]*Dept. of Math. and Life Sciences, Hiroshima Univ.*, [2]*Dept. of Math. and Life Sciences, Hiroshima Univ.*)

Chromosomes consist of various domains with different transcriptional activities separated by insulator sequences. Recent studies suggested that the chromatin loop-forming protein binding sequences represented typical insulators. Alternatively, some long nucleosome-excluding DNA sequences were also reported to exhibit insulator activities without specific bindings of loop-forming proteins. However, the mechanism of the insulator activities of these sequences and the possibilities of similar insulators existing in other organisms remained unclear. In this presentation, we show some results of our recent analysis for these issues by coarse-grained molecular dynamics simulations and genome-wide epigenome data analysis of human cell.

1SEA-5 (1Pos239) Molecular Dynamics of Nucleosome Assembly

Giovanni Brandani, Shoji Takada, Cheng Tan (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

The dynamic assembly of nucleosomes underlies the regulation of epigenetic patterns throughout genomes. We employed molecular dynamics simulations to extensively characterize the environmental factors modulating the kinetics of nucleosome assembly. Markov state modeling reveals the existence of a rich set of metastable partially assembled nucleosome structures, and that specific DNA bending motifs facilitate assembly by directing the binding of histones toward their target. We further show how the flexibility of nucleosomal DNA across yeast promoters is related to both nucleosome occupancy and gene expression, highlighting that sequence can directly contribute to chromatin organization by either facilitating or inhibiting nucleosome assembly at specific genomic regions.

1SEA-6 (1Pos088) エピジェネティック修飾をもつクロマチンのモデルにおける不連続相転移
(1Pos088) Discontinuous Phase Transition in a Chromatin Model with Epigenetic Modification

Kyosuke Adachi, Kyogo Kawaguchi (*RIKEN BDR*)

Chromatin is a complex of DNA and histone proteins. Histones are amenable to several kinds of epigenetic modifications, e.g., acetylation and methylation, which play important roles in regulating the gene expression. Interestingly, recent experiments have suggested megabase (Mb)-scale change in both the chromatin spatial structure and the histone modification profile through differentiation. However, the mechanism of such large-scale change has not been clarified. In this study, we propose a polymer model to describe a chromatin with dynamic histone modifications and find that the model can show a discontinuous phase transition. We discuss the possibility that this phase transition is the physical origin of the Mb-scale chromatin change observed in experiments.

1SEA-7 Transcription dynamics of DNA at interfaces

Tetsuya Yamamoto (*Nagoya Univ., Dep. of Mat. Phys.*)

The dynamics of gene expression is regulated by the structure and dynamics of chromatin. It is of interest to treat the structure and dynamics of chromatin by using an extension of the soft matter physics to study the physical mechanism of the gene regulation. We here use a DNA brush, where DNA is end-grafted to a surface, as a model system of chromatin in a nucleus to theoretically predict i) the dependence of the transcription rate on the packing density of DNA and the orientation of transcription units, ii) the phase separation driven by transcription dynamics, and a part of our current project on iii) the transcription dynamics enhanced by the chromatin looping at interfaces.

1SEA-8 クロマチンの高次構造とダイナミクス ～高分子物理の視点から
Structure and dynamics of chromatins: perspective from polymer physics

Takahiro Sakaue[1,2] ([1]*Department of Physics and Mathematics, Aoyama Gakuin University*, [2]*JST, PRESTO*)

Many observations suggest that the structure and dynamics of chromatin are closely related to the mechanism of gene regulation. Among various aspects of chromatin, the most primal is its very thin and tremendously long thread-like structure. A natural question then arises: Is polymer physics useful here? Recent molecular/cellular biological studies seem to be evidencing that the answer is positive, and at the same time, providing a fundamental challenge to be answered by polymer physics. In the presentation, I will talk about topological effect on the conformation and organization of long chromatin polymers, and the anomalous dynamics of genetic loci. Both topics are relevant to live chromatins in nucleus, and also deeply related to fundamentals in polymer physics.

1SFA-1 Genetically encoded tools for brain sciences

Atsushi Miyawaki (*RIKEN*)

Over the past two decades, various genetically encoded probes have been generated using fluorescent/bioluminescent proteins, and are being used to investigate the function of specific signaling mechanisms in synaptic transmission, integration, and plasticity. I will discuss how the probes have advanced our understanding of the spatio-temporal regulation of biological functions inside cells, neurons, embryos, and brains. I will speculate on how these approaches will continue to improve due to the various features of fluorescent/bioluminescent proteins that serve as the interface between light and life. Due to recent remarkable progress in gene transfer techniques, the experimental animals to be studied are not limited to mice but extended to primates. Newly emerging tools will stimulate the imagination of many scientists, and this is expected to spark an upsurge in the demand for them.

1SFA-2 蛍光全脳イメージングのための連続断層イメージング法 FAST
Block-face serial microscopy tomography for whole-brain fluorescence imaging

Kaoru Seiriki[1,2], Hitoshi Hashimoto[1,3,4,5] ([1]*Lab. Mol. Neuropharmacol., Grad. Sch. Pharmaceutical Sci., Osaka Univ.*, [2]*Interdisciplinary Program for Biomedical Sci., Inst. Transdisciplinary Graduate Degree Programs, Osaka Univ.*, [3]*Mol. Res. Cent. Children's Mental Development, United Grad. Sch. Child Development, Osaka Univ.*, [4]*Div. Biosci., Inst. Datability Sci., Osaka Univ.*, [5]*Transdimensional Life Imaging Div., Inst. Open and Transdisciplinary Res. Initiatives, Osaka Univ.*)

Brain functions are specialized and integrated in different parts of the brain, and therefore imaging the whole brain is important for precise understanding of anatomical and functional brain networks. For this purpose, we have developed a fluorescence microscopic apparatus for whole-brain imaging at a subcellular resolution, named FAST (block-face serial microscopy tomography). FAST is applicable to variety of chemically and genetically labeled brain samples including immediate early gene-reporter mouse brains, which allows brain-wide activation mapping at a cellular level. Hypothesis-free analysis of brain-wide neuronal activation will open new avenues for understanding how various brain cells interact with each other across brain regions for brain function.

1SFA-3 非回折と自己湾曲特性を用いた光ニードル顕微鏡における 3 次元イメージング
Three-dimensional imaging in light needle microscopy utilizing non-diffraction and self-bending characteristics

Yuichi Kozawa, Shunichi Sato (*IMRAM, Tohoku UNiv.*)

The acquisition time reduction for three-dimensional (3D) imaging is highly demanded in many fields, including life science. In laser scanning fluorescence microscopy, in general, 3D image is constructed by two-dimensional image stacking by changing the observation plane, ultimately limiting the acquisition speed. Here we present a new method enabling 3D image acquisition without moving the observation plane in a two-photon excitation microscope employing a light needle spot with an extended focal depth. The depth information is retrieved by utilizing non-diffracting and self-bending characteristics imposed on fluorescent signals. Thus, 3D images can be constructed from a single raster scan of the light needle, which will significantly reduce the acquisition time.

1SFA-4 広視野 2 光子デジタル走査ライトシート顕微鏡とメダカ胚全身イメージングへの応用
Wide-field 2-photon light-sheet microscopy and its application to whole body imaging of medaka embryos

Takashi Saitou, Takeshi Imamura (*Ehime University*)

Light-sheet microscopy provides a suitable way for 4D live imaging of fluorescently-labeled biological specimens at high spatiotemporal resolution and with low phototoxicity. We developed wide-field 2-photon digital scanned light-sheet microscopy by employing near-infrared Bessel beam for illumination optics. Consequently, our optical system achieved ~1.5 mm field of view with ~2 um axial resolution, which makes it possible to perform whole body imaging of Medaka embryos in a single cell resolution. We applied this system to analyze lymphatic vessel formation and intercellularly propagating Ca2+ wave of the developing embryos, which allowed us to detect unique features of both processes. Our system can be a powerful tool for analyzing larger biological samples.

1SFA-5 線虫の全脳機能的イメージングのための要素技術と全神経活動の解析
Bio-image informatics for whole brain activity imaging and analysis of neural activity of C. elegans

Yu Toyoshima[1], Stephen Wu[3], Manami Kanamori[1], Hirofumi Sato[1], Moon Sun Jang[1], Yuko Murakami[2], Suzu Oe[2], Terumasa Tokunaga[4], Osamu Hirose[5], Sayuri Kuge[2], Takayuki Teramoto[2], Yuishi Iwasaki[6], Ryo Yoshida[3], Takeshi Ishihara[2], Yuichi Iino[1] ([1]*Dept of Biological Sciences, Grad Sch of Science, Univ of Tokyo*, [2]*Dept of Biology, Facl of Sciences, Kyushu Univ*, [3]*Inst of Statistical Mathematics, Research Organization of Information and Systems*, [4]*Dept of Systems Design and Informatics, Facl of Computer Science and Systems Engineering, Kyushu Inst of Technology*, [5]*Facl of Electrical and Computer Engineering, Inst of Science and Engineering, Kanazawa Univ*, [6]*Dept. of Mec. Eng., Grad. Sch. of Sci. and Eng., Ibaraki Univ.*)

Recent advances in microscopy techniques enable whole-brain activity imaging in living animals including worms. C. elegans is suitable for neuroscience at network levels because all 302 neurons and their structural connectivity were identified. We obtained time-lapse 3D images of head region of living worms. The neurons were accurately detected, tracked and the neural activities were measured by image analysis methods we developed. Annotating neuron identity is required to compare neural activities between different animals. We developed a semi-automated annotation method that integrated with whole-brain activity imaging. Through analyzing the whole-brain neuronal activity data, we try to elucidate the mechanisms of the neural information processing.

1SFA-6 Bilateral Domain 画像処理
Bilateral Domain Image Processing

Shin Yoshizawa (*IPRT, RAP, RIKEN*)

Advances in imaging technology make us to obtain various biological and physical data as images. Thus, image processing and analysis become important in biophysics. In this talk, a concept of bilateral domain image processing is presented along with our novel applications including edge-aware filtering, enhancement, geometric feature extraction and shape modeling from images. Our bilateral domain consists of coordinates and colors, and an image forms a discrete sampling of manifold in the domain. Our applications are based on anisotropic and geodesic distances on the manifold as well as curvatures of the hyper surface.

1SGA-1 Thick filament activation through a molecular-based mechanosensing, regulates forces in mathematical models of trabecula and ventricle

Lorenzo Marcucci[1,3], Takumi Washio[2], Toshio Yanagida[3] ([1]*Department of Biomedical Sciences, Padova University, Italy*, [2]*Graduate School of Frontier Sciences, The University of Tokyo, Japan*, [3]*Center for Biosystems Dynamics Research, RIKEN, Japan*)

A recent discovery in the regulation of muscle contraction, associates to the classical calcium-mediated thin filament activation, a tension-mediated thick filament activation. In this activation, myosin motors switch between an OFF, or super-relaxed, state, where actomyosin interaction is prevented by their folded configuration along the thick filament, and an ON, or active, state, where a more perpendicular configuration is assumed, toward the thin filament, allowing contraction. The switching rates depend on the tension sustained by the thick filament. This mechanosensing mechanism at the molecular level is associated, through our in-silico methods, to the macroscopic muscle behaviour, as the length-dependent activation in cardiac muscle and the Frank-Starling law.

1SGA-2 (1Pos108) 心筋細胞に備わる収縮リズム恒常性の分子機構の解明
(1Pos108) Elucidation of molecular mechanism of contraction rhythm homeostasis in cardiac myocytes

Seine Shintani[1], Takumi Washio[2] ([1]*Department of Biomedical Sciences, College of Life and Health Sciences, Chubu University*, [2]*Graduate School of Frontier Sciences, the University of Tokyo*)

We found that when the cardiomyocytes are warmed using the an infrared laser, the sarcomere inside the living cardiomyocytes contracts and oscillates only while warming. We named this contractive oscillation as Hyperthermic Sarcomeric Oscillations (HSOs). We considered that the properties of HSOs are likely to be important for the heart. To understand the molecule dynamics, we analyzed the sarcomeric oscillations. As a result, in spite of the large amplitude changes of the oscillations by the calcium concentration, the frequency at each period of HSOs was kept constant. That is, HSOs are equipped with Rhythmic Homeostasis. By the multi-scale cardiomyocyte simulation model, we succeeded in reproducing the HSOs with the presence of myosin and sarcomere populations.

1SGA-3 DNA ナノデバイスと高解像 1 分子イメージング技術を活用した心臓のメカノバイオロジー
Mechanobiology of Heart Revealed with DNA Nano-device and Nanometer-precision Single-molecule Imaging

Mitsuhiro Iwaki[1,2] ([1]*RIKEN, BDR*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*)

In heart, mechanical forces generated by cardiac myosins regulate efficient and cooperative action between mechanosensory proteins, therefore, perturbation of the forces results in heart disease. To clarify the relationship between force and cooperative action, high-resolution imaging of mechanical force and the control should be powerful, however, it is still limited. We recently developed DNA nano-device called “Nanospring”. This is a coil-shaped nanostructure with high brightness, high-resolution force sensing and tunable spring constant. Here, we aim at incorporating nanospring into cardiomyocyte to measure force. Also, we developed a DNA-based molecular platform for dissecting single sarcomeric protein dynamics (e.g. myosin) in the molecular assembly.

1SGA-4 Myosin filament regulation by mechanosensing in skeletal and cardiac muscle

Vincenzo Lombardi (*PhysioLab*)

Contraction of skeletal and cardiac muscles is controlled by a dual filament regulation system. Beyond the classical regulation consisting in Ca2+-induced structural changes in the regulatory proteins on the thin, actin containing, filament promoting the release of the actin sites for binding of myosin motors, a second regulation mechanism, based on the mechano-sensitivity of thick, myosin containing, filaments has been recently defined by X-ray diffraction (Linari et al, 2015 Nature 528, 276; Reconditi et al, 2017 PNAS USA 114, 3240). Thick filament mechanosensing represents an energetically well suited mechanism by which myosin motors are switched ON from their OFF state, in which ATP hydrolysis is inhibited, in relation to the load of the contraction.

1SGA-5 骨格筋再生における機械受容イオンチャネル PIEZO1 の役割
Role of a mechanosensitive cation channel PIEZO1 in skeletal muscle regeneration

Yuji Hara[1,2], Kotaro Hirano[1], Seiji Takabayashi[1], Masaki Tsuchiya[1], Masato Umeda[1] ([1]*Graduate School of Engineering, Kyoto University*, [2]*AMED PRIME*)

Skeletal muscle possesses a highly adaptive and robust capacity to regenerate from degenerative injuries. Regeneration of myofibers largely depend on resident stem cells known as muscle satellite cells (MuSCs), but the mechanisms underlying satellite cell activation remains unclear. Here we show that PIEZO1, a mechanosensitive cation channel that is activated by membrane tension, plays a role in activation of satellite cells. Regeneration of myofibers after cardiotoxin-induced injuries was significantly delayed in MuSC-specific Piezo1-deficient mice. Moreover, PIEZO1 is accumulated at the cleavage furrow during cytokinesis of MuSC. Thus, our results suggest that PIEZO1 plays a role in myofiber regeneration by sensing the membrane tension at the cleavage furrow.

1SGA-6 Mechano-protective roles of sugar chain in skeletal muscle

Motoi Kanagawa (*Kobe Univ. Grad. Sch. Med.*)

Skeletal muscle is a dynamic tissue that routinely undergoes cell shortening and generates forces, and thus, it is required to limit mechanical cellular injury and adapt to changing workloads. Dystroglycan connects extracellular matrix and cytoskeletons across the cell membrane via sugar chain that contains a novel post-translational unit ribitol-phosphate (RboP). RboP forms a tandem repeat and functions as a scaffold for the formation of the ligand-binding moiety. Muscle cell membrane in mouse models that lack the RboP shows fragility against physical stress. These findings demonstrate that RboP-containing sugar chain plays an essential role to maintain membrane integrity and prevent physical stress-induced cell damages.

1SGA-7 Lipid bilayer membrane mediated mechanotransduction in vascular endothelial cells

Kimiko Yamamoto[1], Joji Ando[2] ([1]*System Physiology, Graduate School of Medicine, The University of Tokyo*, [2]*Laboratory of Biomedical Engineering, School of Medicine, Dokkyo Medical University*)

Vascular endothelial cells (ECs) play critical roles in regulating a variety of vascular functions in response to hemodynamic forces, namely, shear stress and stretch. Endothelial plasma membranes have recently been shown to respond differently to shear stress and stretch, by changing their lipid order, membrane fluidity. Artificial lipid-bilayer membranes also show similar changes in the lipid order in response to shear stress and stretch, indicating that these are physical phenomena rather than biological reactions. Such physical changes activate the membrane receptors and cell responses specific to each force. These findings suggest that the plasma membranes of ECs act as mechanosensors with modification of the conformation and functions of the membrane proteins.

1SGA-8 (1Pos268) 細胞内動態をサブセルレベルで制御する温和な NanoHeating 技術
(1Pos268) A Thermodynamic Tool for Mechanobiology Research: Mild Nanoheating Technology to Alter Subcellular Dynamics

Satoshi Arai[1], Nandus Ferdi[2] ([1]*Res. Inst. Sci. Eng., Waseda Univ.*, [2]*WABIOS*)

Recent years, we developed the nanoheating technology that enables to heat up the intracellular local place using a photothermal dye-based nanoparticle with a near infrared laser. Previously, we demonstrated our technology in the induction of apoptosis in cancer cells under harsh elevated temperature. Here, we report several applications using more “mild” heating to change cellular activities. For example, we showed that mild heating could induce the skeletal muscle contraction based on the alteration of protein-protein interaction. In the other case, we found that mild heating affected the dynamics of intracellular ATP. We do believe our methodology will be helpful to provide the viewpoint of thermodynamics to mechanobiology research.

1SHA-1 高圧力による生きた細胞内の分子機械の活性化
Activation of molecular machinery in living cells using high-pressure techniques

Masayoshi Nishiyama (*Kindai Univ.*)

Hydrostatic pressure is an isotropic mechanical action to change the intermolecular interactions between protein and water molecules. This means that applied pressure can induce the structure and function of molecular machines. We developed a high-pressure microscope that is optimized both for the best image formation and for the stability to hydrostatic pressure up to 150 MPa. The developed system have allowed us to monitor the molecular machineries in vitro and in vivo. Here, we show several successive examples to activate their motility at high-pressure. For example, high hydrostatic pressure restores the rhythmical beating motion of paralyzed-flagella mutants of *Chlamydomonas*.

1SHA-2 (1Pos147) RhoA activation induces cell cycle exit and differentiation of skin cancer cells

Oleg Dobrokhotov, Masahiro Sokabe, Hiroaki Hirata (*Nagoya Univ., Grad. Sch. Med.*)

Contact inhibition of proliferation (CIP) is crucial for tissue homeostasis, while loss of CIP is a typical hallmark of cancer. Previously we have shown that actomyosin contractility is required for CIP in HaCaT keratinocytes and pharmacological activation of the RhoA-ROCK-myosin II axis in A431 skin epidermoid carcinoma cells causes inhibition of their proliferation. In the present study, further analyses reveal that activation of RhoA in A431 cells induces cell cycle exit into G0 quiescence, which is followed by their differentiation into cytokeratin-10-positive keratinocytic cells. While cancer stemness drives tumor development, induction of cancer cell differentiation through the activation of RhoA-ROCK signaling might provide a novel strategy for cancer therapy.

1SHA-3 (1Pos146) Direct observation of cell mechanics under high hydrostatic pressure

Masatoshi Morimatsu, Keiji Naruse (*Grad. Sch. of Med., Dent. and Pharma. Sci., Okayama Univ.*)

Activities of daily living such as walking and mastication pressurize tissues. However, the effect of pressurization on cells at a molecular level is poorly understood due to the lack of methods that observe cells under high pressure condition. Here we used hydrostatic pressure microscope to apply high pressure to cells in vitro and investigated cellular morphology, molecules, and gene expression levels. We observed higher pressure decreased the size of cells and gene expression levels of extracellular matrix proteins were increased after pressurizing process. Our results suggest that pressurization to cells is related to maintenance of tissue function.

1SHA-4 (1Pos253) 高圧力下で早くなるシアノバクテリアの概日周期
(1Pos253) Pressure accelerates the circadian clock of cyanobacteria

Ryo Kitahara[1], Katsuaki Oyama[2], Takahiro Kawamura[2], Keita Mitsuhashi[2], Soichiro Kitazawa[1], Kazuhiro Yasunaga[1], Natsuno Sagara[1], Megumi Fujimoto[2], Kazuki Terauchi[2] ([1]*Pharm. Sci., Ritsumeikan Univ.*, [2]*Life Sci., Ritsumeikan Univ.*)

The oscillation period lengths of circadian clocks in diverse organisms are mostly independent of ambient temperature, known as temperature compensation. However, nothing has been known about pressure effects on circadian clocks. Here we report pressure effects on the cyanobacterial circadian clock, consisting of KaiA, KaiB and KaiC proteins. We found that the cycle of the KaiC phosphorylation was accelerated from 22 hours at 1 bar to 14 hours at 200 bar. We also found that ATPase activity measured in kcat of KaiC at 200 bar was 1.5 times greater than that at 1 bar. These results match the proposed correlation between the circadian period length and the ATPase activity of KaiC period-mutant proteins.

1SHA-5 細菌べん毛モーター回転に及ぼす高静水圧の影響
Effects of high hydrostatic pressure on the rotation of the bacterial flagellar motor

Ikuro Kawagishi[1,2] ([1]*Dept. Frontier Biosci., Hosei Univ.*, [2]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

The flagellar motor of *Escherichia coli* is an elaborate molecular machine that transduce proton motive force into mechanical force to rotate the axial structure consisting of the rod, the hook and the filament. One remarkable feature of the motor is that it can instantaneously switch directional sense without changing the direction of proton flow. Whereas the motor rotates counterclockwise (CCW) without any stimuli, clockwise (CW) rotation is induced upon binding of the phosphorylated form of the response regulator CheY (CheY-P). We have demonstrated that high hydrostatic pressure can induce CCW rotation even in the presence of CheY-P. This enabled us to analyze motor switching *in vivo* by modulating the CheY-motor interaction with altered hydrostatic pressure.

1SHA-6 圧力感受性変異 YFP の圧力応答の構造基盤
Structural basis of pressure response of a pressure sensitive YFP variant protein

Katsumi Imada[1], Mika Tsujii[1], Takayuki Nagae[2], Hiroaki Hata[3], Tomonobu Watanabe[4], Masayoshi Nishiyama[5], Akio Kitao[3], Nobuhisa Watanabe[2] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Eng., Nagoya Univ.*, [3]*Sch. LifeSci and Tech,. Tokyo Inst. Tech.*, [4]*BDR, Riken*, [5]*Sch.Sci. and Eng., Kindai Univ.*)

Yellow fluorescent protein (YFP) based sensors are widely used to detect intracellular information. We recently found that a YFP mutant protein with insertion of three glycine residues at β7 (YFP-3G) changes its fluorescence depending on hydrostatic pressure. To understand the molecular mechanism of the fluorescence change by pressure, we determined crystal structures of YFP-3G and measured the fluorescence spectra of the crystals under high and low pressures. We found that the chromophore conformation is flipped and the disordered part of β7 becomes ordered at high pressure. MD simulation reproduced the structural changes. These structural changes may cause the fluorescence response of YFP-3G to pressure.

1SHA-7 Pressure effects on protein-protein interactions studied by molecular dynamics simulations

Hiroaki Hata[1], Yasutaka Nishihara[2], Masayoshi Nishiyama[3], Ikuro Kawagishi[4], Akio Kitao[1] ([1]*Dept. of Life Sci. and Tech., Tokyo Tech*, [2]*IMCB, UTokyo*, [3]*Grad Sch. of Sci. and Eng., Kindai Univ.*, [4]*Dept. of Frontier Biosci., Hosei Univ.*)

Protein-protein interactions are essential for functional regulation of biomolecules. However, it currently needs a very long computational time to simulate association/dissociation between proteins using conventional molecular dynamics (MD) methods. Here we simulated a dissociation of a protein complex, CheY-FliM, which plays a key role in bacterial chemotaxis, using the efficient conformation sampling method, parallel cascade selection molecular dynamics (PaCS-MD), and investigated the pressure effect on the interactions. We found that the obtained binding free energy decreased as pressure increases, mainly due to enhanced protein hydration induced by pressure.

1SBP-1 Microscopic measurements of force and taxis in bacteria/archaea

Takayuki Nishizaka, Daisuke Nakane (*Dept. Phys., Gakushuin Univ.*)

An optical microscope is a powerful tool to quantify variety of unknown behaviors of microorganisms such as bacteria and archaea with sizes of microns. In addition to their collective motions, single-steps of their motor elements were revealed in detail under advanced light-illumination on the ultra-stable sample stage (Kinosita... & Nishizaka, *PNAS* 2014; Iwata... & Nishizaka, *Commun. Biol.* 2019). We've recently focused on the phototactic regulation of machinery that induces twitching motility (Nakane & Nishizaka, *PNAS* 2017), and here report the new aspect of taxis; the rheotaxis in parasitic *Mycoplasma pneumoniae* and *Mycoplasma mobile* under controlled fluid flow rate. The possible mechanism underlying how they change geometrical orientations will be investigated.

1SBP-2 Biophysical models of physical rupturing of bacterial cells by nano-structured surfaces

Elena Ivanova (*RMIT University*)

The threat of a global rise in the number of untreatable infections caused by antibiotic resistant bacteria calls for the design of a new generation of bactericidal materials. The nano-pattern on the surface of Clanger cicada (*Psaltoda claripennis*) wings represents the first example of a new class of biomaterials that can kill bacteria on contact based solely on its physical surface structure. We developed the first biophysical model based on the bactericidal action of cicada wings to provide increased fundamental understanding of the mechanisms behind this recently discovered phenomenon. This work highlighted that the high bactericidal activity of high aspect ratio nanofeatures can outperform both natural bactericidal surfaces.

1SBP-3 心筋ナノイメージング
Cardiac nano-imaging: from cells to the heart

Norio Fukuda (*Department of Cell Physiology, The Jikei University School of Medicine*)

To analyze cardiac excitation-contraction coupling, we applied nanometry techniques to cardiology. We found that in cardiomyocytes, when Ca2+ transients are synchronized along a myofibril, the averaging of sarcomere length (SL) causes a marked underestimation of the magnitude of displacement due to individual sarcomeric behaviors. Then, we developed a confocal-imaging system for the beating heart in anesthetized mice. As a result, individual SL values varied markedly in the same myocyte during the cardiac cycle and sarcomeres along a myofibril alternately contributed to myocardial dynamics. Therefore, sarcomeres behave distinctly according to the physiologic condition, and mechanical interactions between sarcomeres along a myofibril may organize myocardial dynamics.

1SBP-4 How Japanese researchers can get access failing and donor tissue from the Sydney Heart Bank. A viable alternative to using animal models

Cristobal G. dos Remedios, Amy Li, Sean Lal (*Bosch Institute, Discipline of Anatomy & Histology, University of Sydney*)

Japanese publications suggest there is a real interest in investigating the molecular mechanisms of human heart failure (HF) but few reports employ tissues from failing and healthy human hearts. Most use animal models, particularly mouse. Unfortunately only ~50% of mouse models translate to man. The Sydney Heart Bank has >25,000 (LV, RV, LA, RA) tissue samples from 350 transplantated hearts, and 120 donors that are available for research projects. Diseases include idiopathic and familial dilated cardiomyopathies, hypertrophic CM, peripartum CM, and end-stage ischemic heart disease. All samples are snap frozen in liquid N within minutes to avoid post-mortem artefacts and can be delivered to Japan at 180°C. In our 130 publications Japanese labs are under-represented.

1SBP-5 リン脂質 flippase による細胞膜変形と細胞機能
Plasma membrane deformation by phospholipid flippase and cellular functions

Hye-Won Shin (*Grad Sch Pharm Sci, Kyoto Univ*)

In eukaryotic cells, the lipid bilayer of the plasma membrane exhibits asymmetric lipid distributions. Aminophospholipids are limited in the cytoplasmic leaflet whereas phosphatidylcholine (PC) and sphingomyelin are enriched on the cell surface. P4-ATPases, a subfamily of P-type ATPases, translocate phospholipids from the exoplasmic to the cytoplasmic leaflet of cellular membranes. Among P4-ATPases, we have shown that ATP11A/C translocate aminophospholipids, and ATP8B1/2, and ATP10A translocate PC specifically. We also showed that the exogenous expression of PC-flippase changed cell shapes and was able to drive membrane deformation. Here, I would like to discuss membrane deformation and cellular functions in connection with PC-flippase.

1SBP-6 Structural definition of phospholipid-mediated oligomerization of defensins in fungal and tumour cell lysis

Marc Kvansakul, Mark Hulett, Sofia Caria, Ivan Poon, Michael Jarva, Kha Tran Phan, Fung Lay, Amy Baxter (*La Trobe University*)

Cationic antimicrobial peptides (CAPs) such as defensins are ubiquitously found innate immune molecules that often exhibit broad activity against microbial pathogens and mammalian tumour cells. Many CAPs act at the plasma membrane of cells leading to membrane destabilization and permeabilization. Here we describe a novel cell lysis mechanism for fungal and tumour cells by plant and human defensins that act via direct binding to the plasma membrane phospholipids. We have determined the crystal structures of the plant defensins NaD1 and NsD7, revealing distinct oligomeric arrangements. Our observations identify a conserved innate recognition system by defensins for direct binding of phospholipid that permeabilize cells via a novel membrane disrupting mechanism.

1SCP-1 Simultaneous spatio-temporal dendritic voltage/calcium mapping and somatic recording from Purkinje neurons in awake mice

Bernd Kuhn, Christopher J. Roome (*OIST Graduate University*)

Spatio-temporal maps of dendritic signalling is fundamental to neuronal information processing, yet remain unexplored in awake animals. We combine simultaneous sub-millisecond voltage and calcium two-photon imaging from distal spiny dendrites, with somatic electrical recording from spontaneously active cerebellar Purkinje neurons (PN) in awake mice. We detect discrete 1-2 ms supra-threshold voltage spikelets in the distal spiny dendrites during dendritic complex spikes. Spikelets and their calcium correlates are highly heterogeneous in number, timing and spatial distribution within and between complex spikes. Highly variable 5-10 ms voltage hotspots represent postsynaptic potentials, allowing for the first time mapping of synaptic input onto a neuron in an awake animal.

1SCP-2 Novel "*in vivo*" two-photon microscopy for vast and longtime neural activity

Tomomi Nemoto (*RIES, Hokkaido Univ.*)

in vivo" two-photon microscopy has become widely used in neuroscience. By a newly-developed 1064 nm laser-diode-based light source, we visualized pyramidal neurons in the hippocampal dentate gyrus layer without removing any cortical layers above hippocampus (*Biomed. Opt. Express*, 2015). Noticeably, we sucessfully demonstrated video-rate Ca^{2+} imaging of hippocampal CA1 pyramidal neurons expressing a new Ca^{2+} indicator, XCaMP (*Cell*, 2019). Also, we proposed an optical brain clearing method for "*in vivo*" imaging, MAGICAL (*BioRxiv.*, 2019), that enabled to capture vivid images of hippocampal CA1 neurons at fast speed. In this presentation, we will discuss improvements and future applications based on recent data.

1SCP-3 機能的光干渉断層法とフーリエイメージングによる脳機能構造の 3 次元マイクロ計測
Functional optical coherence tomography with Fourier imaging reveals three-dimensional and micro-scale brain functional structure

Yu Nakamichi, Manabu Tanifuji (*RIKEN CBS*)

Optical coherence tomography (OCT) is a technique to visualize 3D structure of biomedical tissue with micro-scale spatial resolution, and has been applied to functional imaging (functional OCT; fOCT) where small changes of light scattering elicited by neural responses are detected as functional signals from the brain. However, it was difficult to clearly map 3D functional structure due to physiological noises. Here, we combined fOCT with Fourier imaging that enables to remove such noises by analyzing temporal responses to periodic stimulation. We demonstrated this technique in cat early visual cortex by visualizing 3D structures of orientation columns and validating the structures with electrophysiological recordings.

1SCP-4 偏光で解き明かす生細胞内分子アセンブリーのナノ構造とそのダイナミクス
Dissecting nano-scale architectures and dynamics of molecular assemblies in living cells with polarized light

Tomomi Tani (*Marine Biological Laboratory, Woods Hole*)

Spatial and temporal regulation of orders in molecular assemblies drives the fundamental functions of living cells. We developed a novel fluorescence polarization microscope that reports the molecular orientation of biological molecules in 3D in living cells with single molecule sensitivity. We mapped the orientation of F-actin networks at the leading edge of migrating human skin cells, revealing the architectural dynamics of actin networks that relate to cell migration. The system has also used to explore the structures of membrane receptors integrins in migrating cells upon interacting with extracellular matrices. I will introduce some trials to observe activity dependent architectural changes of neuronal cells in mouse hippocampus using polarized light.

1SCP-5 (1Pos005) The role of C-terminal carboxylation in α-conotoxin LsIA interactions with human α7 nicotinic acetylcholine receptor *in silico*

Jierong Wen, Andrew Hung (*Sch. Sci., RMIT Univ.*)

Nicotinic acetylcholine receptors (nAChRs) are ligand-gated ion channels, modulating the signal transmission between neurons. However, their subtype-specific functions are still unclear due to the lack of suitable selective probes. α-Conotoxins are well-known to inhibit nAChRs with high potency, nevertheless, the knowledge of the effects of C-terminus (C-T) modification on subtype selectivity is sparse. In this study, we identified the molecular determinants of α-conotoxin LsIA, with amidated/ carboxylated C-T, binding to human α7 nAChR via molecular dynamics simulations. Results show the atomic-level mechanism of reduced carboxylated LsIA affinity at the α7 versus the amidated LsIA, which may open a new avenue for the design of new leads for nAChR-related diseases.

1SCP-6 (1Pos266) グルタミン酸受容体を介した植物の長距離 Ca^{2+}シグナル
(1Pos266) Long-distance Ca^{2+} transmission via glutamate receptor channels in plants

Masatsugu Toyota[1,2] ([1]*Dept Biochem and Mol Biol, Saitama Univ*, [2]*University of Wisconsin-Madison*)

In the vertebrate central nervous system, glutamate acts as an excitatory neurotransmitter, regulating synaptic transmission via activation of glutamate receptor channels and facilitating long-distance information exchange throughout their bodies. Similarly, plants sense local stresses, such as insect attack, and transmit this information throughout their bodies to rapidly activate defence responses in undamaged organs. Here we show that glutamate is a wound signal in plants that leaks from damaged cells and activates the *GLUTAMATE RECEPTOR LIKE* family of ion channels in *Arabidopsis*, resulting in the intracellular Ca^{2+} transmission to distant organs (Toyota et al Science 2018). These data help understand long-range molecular signaling networks in animals and plants.

1SCP-7 光信号で「見る」神経回路のはたらき–膜電位感受性色素（VSD）を中心に
Optical view of the brain neural circuit activity: Voltage-sensitive-dye (VSD) imaging

Takashi Tominaga, Yoko Tominaga (*Inst. Neurosci., Tokushima Bunri Univ.*)

Voltage-sensitive dye (VSD) was developed to acquire fast membrane potential changes in the excitable membrane using optical measurement devices as early as the 1970s. Soon, developments of large-scale imaging device enable us to visualize the activity of the neural circuit. However, the simple wide-field VSD imaging methods have long been limited technique for some limited laboratories until the late 1990s, mostly because of its poor signal noise ratio (SNR). Here, we will introduce how we solved the technical difficulties, and the VSDI became to be a "conventional" method. I will show how we can use the technique to address the functions of brain slice preparation. Also, we will introduce a new kind of fast intrinsic optical signal (FIOS).

1SDP-1 溶液中での蛋白質間相互作用の定量的解析法
Quantitative assessments of intermolecular protein mediated interactions in solution

Susumu Uchiyama[1,2] ([1]*Department of Biotechnology, Graduate School of Engineering, Osaka University*, [2]*ExCELLS*)

Quantitative assessments of intermolecular protein mediated interactions in solution is a key for the understandings of a biological systems from the view points of energetics. We have been analyzed protein-protein and protein-ligand interactions in dilute and highly concentration conditions. In this presentation, with principles and actual examples, I'll introduce classical but effective biophysical approaches and recent developed methods for the quantitative inter-molecular analysis in solution.

1SDP-2 Native mass spectrometry of biomolecular complexes

Satoko Akashi (*Grad. Sch. Med. Life Science, Yokohama City Univ.*)

Native mass spectrometry (native MS) enables observation of non-covalently bound complexes. With native MS, we have succeeded in characterization of the protein complexes reconstituted with recombinant proteins, such as nucleosome core particle (204 kDa), which is the minimum structural unit of chromatin. In the present paper, advantages of native mass spectrometry are discussed, and application of native MS to characterization of the biomolecular complexes (reconstituted nucleosome core particle, protein-ligand complexes under crude conditions, etc.) is demonstrated.

1SDP-3 (1Pos055) Biophysical analysis of alpha-synuclein oligomers by microchip electrophoresis

William E. Arter[1,2], Catherine K. Xu[1], Georg Krainer[1], Christopher M. Dobson[1], Tuomas P. J. Knowles[1,2] ([1]*Centre for Misfolding Disease, Department of Chemistry, University of Cambridge*, [2]*Cavendish Laboratory, Department of Physics, University of Cambridge*)

Alpha-synuclein is a protein strongly implicated in neurodegenerative disease, it is known to misfold and self-assemble into insoluble, fibrillar deposits in the brains of Alzheimer's patients. Oligomeric aggregates of synuclein are transient, intermediate and toxic species in this process. Studying them remains a challenge for traditional biophysical techniques due to their high degree of heterogeneity, short lifetime and low abundance. Here, we present microfluidic electrophoresis as a novel technique for the biophysical analysis of oligomeric synuclein. Our method enables the highly sensitive study of transient, heterogenous oligomers in the solution-phase, and provides a platform for further examination of protein complexes and protein-protein interactions.

1SDP-4 (1Pos267) Visualization and quantification of biological samples by high-speed atomic force microscope

Hiroki Watanabe[1,2], Koichi Kato[1,2,3], Takayuki Uchihashi[1,4] ([1]*NINS, ExCELLS*, [2]*NINS, IMS*, [3]*Grad. Sch. Pharm. Sci., Nagoya City Univ.*, [4]*Dept. Phys., Nagoya Univ.*)

High-speed atomic force microscope (HS-AFM) allows us to visualize the dynamic structural changes of biological samples and to intuitively understand their functional mechanism under physiological conditions at high spatiotemporal resolution. Using HS-AFM, recently, not only the direct observation of dynamic conformational change at the single-molecule level but also the quantification of the binding/dissociation process between individual molecules has been carried out. Additionally, we are trying to extend the range of objects by combining with fluorescence microscopy or by obtaining an additional information such as mechanical properties of samples. In this presentation, we will show analytical method of several experiments and discuss about the capability of HS-AFM.

1SDP-5 Dynamic structures and interactions of antibodies under physiologically relevant conditions

Saeko Yanaka[1,2,3], Rina Yogo[1,2,3], Hirokazu Yagi[3], Koichi Kato[1,2,3] ([1]*IMS., Natl. Inst. Nat. Sci.*, [2]*ExCELLS, Natl. Inst. Nat. Sci.*, [3]*Grad. Sch. Pharma. Sci., Nagoya City Univ.*)

Antibodies are glycoproteins having modular structures that possess significant degrees of internal motions on varying spatiotemporal scales. The quaternary-structure plasticity of antibodies is relevant to their versatile intermolecular interactions for expressing immune functions. To gain deeper insights into the functional mechanisms of antibodies, we characterize their dynamic structures and interactions by combining experimental and computational approaches. Furthermore, we characterize the antibody interactions in physiologically more relevant systems typified by blood environments. The NMR spectroscopic techniques we developed offer useful tools for characterizing dynamic behaviors of antibodies under physiological conditions.

1SDP-6 (1Pos058) 新規に開発した高濃度タンパク質のためのネガティブ染色電子顕微鏡法 (1Pos058) A newly developed negative stain EM method for protein complexes at high protein concentration

Hiroshi Imai[1], Takayuki Kato[2], Gerle Christoph[3], Etsuko Muto[4], Kaoru Mitsuoka[5], Genji Kurisu[3], Keiichi Namba[2], Takahide Kon[1] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*IPR, Osaka Univ.*, [4]*CBS, RIKEN*, [5]*Res. Ctr. UVHEM, Osaka Univ.*)

The negative stain EM method is a powerful screening tool to evaluate protein quality prior to cryo-EM. However, the typical protein concentration for negative staining EM is 100 times lower than that for cryo-EM. Proteins weakly associated with a protein complex (e.g. with a Kd in the millimolar range) may fall off by dilution for negative stain EM. Here we have developed a novel negative stain EM method for high concentration protein complexes in order to rapidly visualize a weakly associated protein on a protein complex. We applied this method to glutamate dehydrogenase (GDH), which is known to form a linear polymer only under high concentration. This method has successfully visualized the high-concentration GDH structure. We also observed such structure by cryo-EM.

1SDP-7 蛋白質相互作用の熱測定と創薬
Thermodynamics of Protein Interaction for Therapy and Diagnosis

Satoru Nagatoishi[1,2], **Kohei Tsumoto**[1,2] ([1]*Inst Med Sci, Univ Tokyo*, [2]*Sch Eng, Univ Tokyo*)

Thermodynamic analyses of protein interactions has become one of the fundamental approaches for drug discovery and development, which is due to the progresses on molecular and structural biology. Recently, thermodynamic information is fundamental for compound screening and optimization, and also is essential for screening and optimization of macromolecular drug candidates. Here we summarize the background and current status of thermodynamics-based drug researches and discuss the perspectives.

1SEP-1 Cellomics approach for high-throughput functional annotation of Caenorhabditis elegans neural network

Wataru Aoki[1,2], Yuji Yamauchi[1], Mitsuyoshi Ueda[1] ([1]*Graduate School of Agriculture, Kyoto University*, [2]*JST, PREST*)

One of the primary objectives of neuroscience is to understand how computations are implemented across neural networks. However, even in C. elegans, the relationships between behaviors and neural networks have not been comprehensively revealed. We proposed a novel cellomics approach enabling high-throughput and comprehensive exploration of the functions of single neurons [1]. To realize this, we established a C. elegans library where opsin is labeled in a randomized pattern. Behavioral analysis on this library under light illumination enabled high-throughput annotation of neurons affecting target behaviors. This cellomics approach will lead to the accumulation of neurophysiological and behavioral data of the C. elegans neural network.
[1] Sci Rep, 8, 10380 (2018)

1SEP-2 (1Pos196) Single-cell trajectory analysis of human iPS cell-derived neurons carrying a rare RELN deletion

Yuko Arioka[1,2,3], Emiko Shishido[1,4], Norio Ozaki[1] ([1]*Department of Psychiatry, Nagoya University Graduate School of Medicine*, [2]*Nagoya University Hospital*, [3]*Institute for Advanced Research, Nagoya University*, [4]*National Institute for Physiological Sciences*)

To reach the correct destination, newborn neurons must migrate under strictly controlled conditions. However, the migration mechanisms in human neurons remain poorly understood, because of ethical concerns against the use of living human brain. To address this, we performed live-imaging analysis using human iPS cells-derived neurons. Single-cell trajectory analysis revealed that human neurons possessed directional migration even in vitro and their direction is highly correlated with cell shape orientation. On the other hand, neurons with a rare RELN deletion demonstrated a wandering type of migration and a failure of correlation between migration direction and cell shape. Our findings provide molecular and physical bases of migration mechanisms in human neurons.

1SEP-3 Multiphoton imaging and photostimulation techniques by spatio-temporal control of excitation pulses

Keisuke Isobe, Katsumi Midorikawa (*RIKEN RAP*)

Two-photon excited fluorescence (TPEF) microscopy has become a powerful tool for deep imaging of a biological tissue, though it is intrinsically difficult. Even if we use TPEF microscopy, the out-of-focus background fluorescence limits the fundamental imaging depth. Wavefront distortion, which is caused by the inhomogeneous refractive index distribution of tissues, also makes deep imaging difficult because it results in resolution degradation and signal reduction. In addition, there is a trade-off between the temporal resolution and imaging area. In this presentation, we present the spatio-temporal control of excitation pulses to solve these problems.

1SEP-4 Chemical probes for fluorescence imaging or ablation of lacZ-positive cells with single cell resolution

Mako Kamiya (*Grad. Sch. Med., Univ. Tokyo*)

Selective imaging of targeted cells in living samples with chemical probes remains highly challenging. We have developed precisely functionalized activatable fluorescence probes for β-galactosidase, SPiDER-βGal, to selectively label lacZ-positive cells at single-cell resolution in living organisms or tissues. This design strategy based on intramolecular spirocyclization and quinine methide chemisty, in which fluorescence and biding ability to intracellular nucleophiles are simultaneously activated upon reaction with the enzyme, can be expanded to design a red-shifted activatable fluorescence probe for β-galactosidase or to design an activatable photosensitizer capable of specific ablation of lacZ-positive cells with single-cell resolution.

1SEP-5 三次元バーテックスモデル：三次元多細胞動態の 1 細胞統合モデリング
3D vertex model: single cell-integrated modeling of multi-cellular dynamics in three-dimensions

Satoru Okuda[1,2] ([1]*Nano LSI, Kanazawa Univ*, [2]*JST PRESTO*)

Multi-cellular interaction is a key process of causing macroscopic tissue dynamics such as organogenesis, wound healing, and disease. Because of the complexity of multi-cellular interaction, it is difficult to predict macroscopic tissue dynamics linearly from individual cell behaviors. To overcome this problem, we developed a versatile three-dimensional vertex model. This model describes individual cells behaviors such as cell deformation, movement, division, apoptosis, and transport of signal molecules, and predicts three-dimensional multi-cellular dynamics emerging from multi-cellular interactions. This model can be broadly applied to various multi-cellular dynamics and thus open a new avenue for physical understandings of multi-cellular dynamics.

1SEP-6 分子解像度での生命理解に向けて
Towards molecular-resolved biology

Yuichi Taniguchi (*RIKEN BDR*)

In our PRESTO project, we developed two technologies towards resolving molecular details in biological processes. One is a technology for analyzing nucleosome-level 3D genome structure. This technology couples a sequencing assay with molecular dynamics simulation to reveal 3D spatial distributions of nucleosomes and their orientations across the genome. The results uncovered two types of basic secondary structural motifs in nucleosome folding: α-tetrahedron and β-rhombus. The other is a technology for 3D single molecule imaging for a general use. We recently developed a microscopy to visualize single molecule fluorescence up to a 0.2 mm sample depth on a coverslip. This technology will be useful for not only cell/tissue imaging, but also sensitive biochemistry analysis.

1SEP-7 1 細胞操作のための光応答性細胞固定化剤の開発
Photo-responsive cell immobilization tools for single-cell manipulation

Satoshi Yamaguchi[1,2] ([1]*Research Center for Advanced Science and Technology (RCAST), The University of Tokyo*, [2]*PRESTO, JST*)

Spatiotemporal regulation of cell immobilization has attracted a great deal of attention in wide variety of cell science and engineering fields, from cell-cell communication analysis to cell sorting, tissue engineering and organ-on-a-chip. We are now developing photo-responsive cell immobilization tools to attach and release living cells at the desired timing and location onto/from the substrate. Our synthetic materials could simply and rapidly construct micro-patterns of any types of mammalian cells through light-navigated anchoring onto cell surfaces and dynamically release cells through photo-cleavage or photo-isomerization reactions. On the substrate, a single cell array was simply prepared and applied to high-throughput single-cell analysis.

1SFP-1 単結晶中性子回折計 iBIX の現状と将来展望
Current status and future prospects of single-crystal neutron diffractometer iBIX at pulsed neutron source MLF, J-PARC

Kastuhiro Kusaka[1], Taro Yamada[1], Naomine Yano[1], Takaaki Hosoya[1], Takashi Ohhara[2], Ichiro Tanaka[1] ([1]*Frontier Researc Center for Applied Atomic Sciences, Ibaraki University*, [2]*Japan Atomic Energy Agency, J-PARC Center*)

Single crystal neutron diffraction is one of the powerful tools to provide the structure information of all atoms of protein crystals including the hydrogen atoms. High performance time-of-flight single-crystal neutron diffractometer iBIX (IBARAKI biological crystal diffractometer) was developed at next-generation neutron source MLF, J-PARC to elucidate the hydrogen, protonation and hydration structures of biomacromolecules. At present, MLF is operated at 500kW of accelerator power (Maximum power: 1MW) and then beam time for biomacromolecules by iBIX can be provided stably to users. The current status and future prospects and recent scientific outcomes which make the most of the merit of the neutron diffraction experiment by iBIX will be reported.

1SFP-2 中性子結晶構造解析で明らかになるセルラーゼの加水分解メカニズム
Hydrolytic mechanisms of inverting cellulases clarified by neutron crystallography

Kiyohiko Igarashi[1,2] ([1]*University of Tokyo*, [2]*VTT Technical Research Centre of Finland*)

Neutron crystallography is one of the promising techniques to investigate reaction mechanisms of enzyme because it can visualize hydrogens and protons in protein crystal. In recent years, we have tried use the technique to understand the actual mechanisms of cellulases hydrolyzing cellulose, which is the most abundant biomass on the Earth. We have identified various function of imidic acid form of asparagine in proton relay and catalysis so far, and will discuss new insight in neutron crystallography.

1SFP-3 フェレドキシン依存性ビリン還元酵素の機能と基質のプロトン化状態
Function of a ferredoxin-dependent bilin reductase and the protonation state of its substrate

Masaki Unno[1,2] ([1]*Graduate School of Science & Engineering, Ibaraki University*, [2]*Frontier Research Center for Applied Atomic Sciences*)

Ferredoxin-dependent bilin reductases (FDBRs) are enzymes that catalyze biosynthesis of bilin pigment from biliverdin (BV). PcyA, a FDBR, shows two peaks in visible resion of the absorption spectrum when forming complex with BV. These two peaks are suggested as the two protonation states of the substrate BV. We found some mutants of PcyA changed the spectra significantly when forming complex with BV. We conducted neutron crystallography and other techniques to reveal the protonation states of these mutants. In this conference, we present some results of our research for PcyA and its mutants using some techniques including neutron crystallography and X-ray free electron laser, and also discuss about the relationship in the function and protonation states of PcyA.

1SFP-4 (1Pos021) 創薬標的タンパク質の中性子結晶構造解析
(1Pos021) Neutron crystallographic analysis of drug-target proteins

Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

Neutron crystallography enables the direct visualization of hydrogen/deuterium atoms in proteins and thus is a useful tool for revealing protein-drug interactions in detail. In this decade, we determined the neutron crystal structures of transthyretin (TTR), farnesyl pyrophosphate synthase (FPPS) and bromodomain-containing protein 4 (BRD4). TTR is associated with hereditary ATTR amyloidosis. FPPS and BRD4 are the targets of osteoporosis and anti-cancer drugs, respectively. Based on the neutron structures, we will discuss the molecular stability of TTR associated with amyloidogenesis, the protonation state of the bisphosphonates bound to FPPS and the molecular origin of the binding enthalpy of BRD4 inhibitors.

1SFP-5 Liquid properties and chemical reactions in 100 nm nanochannels

Kazuma Mawatari (*Univ. Tokyo*)

Nanochannels with 100 nm space are gathering interests due to unique liquid properties and chemical reactions in the space. Our group developed fundamental technologies to integrate chemical processes in the nanochannel and revealed many unusual liquid properties. In particular, proton diffusion coefficients were enhanced when the space size was reduced to 100 nm order. In addition, an enzymatic reaction was accelerated without increasing the temperature. In this presentation, we will introduce these unique phonomena in the nanochannels.

1SFP-6 光合成水分解反応におけるプロトンおよび水分子の赤外分光検出
Infrared detection of protons and water molecules in photosynthetic water oxidation

Takumi Noguchi (*Division of Material Science, Graduate School of Science, Nagoya University*)

Infrared spectroscopy is a powerful method to detect protons in proteins. In particular, light-induced Fourier transform infrared (FTIR) difference spectroscopy has been extensively used to investigate the reactions of water molecules, the proton release processes, and the protonation states of amino-acid residues in photosynthetic water oxidation performed in photosystem II. In addition, time-resolved infrared spectroscopy (TRIR) has been used to monitor the electron and proton transfer reactions during intermediate transitions in the water oxidation cycle. These data obtained by FTIR and TRIR studies on the proton and water reactions provide crucial information to understand the molecular mechanism of photosynthetic water oxidation.

1SGP-1 ポピュレーションアニーリングによるベイズ推定
Bayesian inference with population annealing

Koji Hukushima (*The University of Tokyo*)

As the amount of data to be analyzed increases in recent years, the amount of calculation required also increases. The problem of computational complexity is an issue that can not be overlooked, even in the Monte Carlo method, which can be calculated generically as well as complex statistical models. One solution is to use a high-performance computer, but in the age of massively parallel computers, the Markov chain Monte Carlo method based on Markov chains is not necessarily suitable for massively parallel computing. Here, we focus on population annealing that is compatible with parallel calculation and can calculate expected value correctly, evidence, Bayesian factor, and leave-one-out cross validation. Some applications in natural science are discussed.

1SGP-2 Objective and efficient procedure for inferring couplings in neuronal networks

Yu Terada[1,2], Tomoyuki Obuchi[1], Takuya Isomura[2], **Yoshiyuki Kabashima**[1] ([1]*Tokyo Tech.*, [2]*RIKEN CBS*)

Inferring couplings from the spike data of neuronal networks is of great interest in neuroscience. Here, we propose an inference procedure for this goal based on the so-called McCulloch-Pitts neuron model. The procedure is composed of two steps: (1) determination of the time-bin size for transforming the spike data to discrete time binary data and (2) screening of relevant couplings from the estimated networks. For these, we develop simple methods based on information theory and computational statistics. Applications to data from artificial and in vitro neuronal networks show that the proposed procedure performs fairly well when identifying relevant couplings, including the discrimination of their signs, with low computational cost.

1SGP-3 定量的安定同位体標識とテンソル分解による重複 NMR シグナルの分解法
Solving signal overlap in NMR spectra using quantitative isotope labeling and tensor decomposition

Takuma Kasai[1,2], Shunsuke Ono[2,3], Toshiyuki Tanaka[4], Shiro Ikeda[5], Takanori Kigawa[1,3] ([1]*RIKEN BDR*, [2]*PRESTO, JST*, [3]*Sch. Comput., Tokyo Inst. Tech.*, [4]*Grad. Sch. Inform., Kyoto Univ.*, [5]*Inst. Stat. Math.*)

Signal overlap is often an obstacle to protein analysis by NMR. Here we report SiPex (Stable-isotope-assisted Parameter extraction), a novel method to solve signal overlap using amino-acid selective isotope labeling and tensor decomposition. We applied SiPex to ^{15}N relaxation measurements. Differences in signal intensities among samples not only help signal assignments by providing amino acid information but also give a clue to separation of overlapped signals. Because each decomposed component contains both information of amino acid and relaxation, SiPex allows us to skip chemical shift determination, which has been required to identify signals in relaxation spectra. This alternative way of protein analysis will expand capability of NMR to analyze difficult targets.

1SGP-4 高速高精度一分子計測により明らかとなったキチン分解酵素の運動機構
Moving mechanism of chitin hydrolase was revealed by high precision and speed single molecule analysis

Akihiko Nakamura[1,2], Kei-ichi Okazaki[1], Tadaomi Furuta[3], Minoru Sakurai[3], Ryota Iino[1,2] ([1]*Institute for Molecular Science*, [2]*SOKENDAI*, [3]*Tokyo Institute of Technology*)

SmChiA is a chitin hydrolase moves on a chitin crystal with hydrolysis of the molecular chain without ATP. To reveal the moving mechanism of SmChiA, we resolved the 1 nm stepping motion by labeling with gold nano probe. Results of stepping ratio between forward and backward steps and time constituents of catalysis and backward step indicated SmChiA biased forward due to the destabilization of backward state after hydrolysis of chain. In the molecular dynamics simulation, chitin chain moved backward and forward without any force from sliding intermediate X-ray structure. These results indicated SmChiA is a Burnt-bridge Brownian ratchet motor.

1SGP-5 DNA curtains: high-throughput single molecule imaging for DNA transactions

Tsuyoshi Terakawa (*Grad. School of Sci., Kyoto Univ.*)

Essential genomic information is written in DNA. DNA binding proteins govern read-out (transcription), preservation (repair), and regulation (chromatin conformation change) of the information. Thus, revealing molecular mechanisms of various DNA binding proteins are expected to shed light on the foundation of life. One of the powerful methods to accomplish this is to observe purified DNA binding proteins on DNA directly. We have observed the DNA binding proteins at single molecule level with our original fluorescent microscopic method called "DNA curtain." I will show a couple of application examples of this technique and discuss the possible extension using simulation-data-driven approaches.

1SGP-6 2 次元／3 次元 AFM によるバイオ系試料の観察と高度な AFM データ解析の必要性
2D/3D-AFM imaging of biological systems and demands for advanced AFM data analysis

Takeshi Fukuma (*NanoLSI, Kanazawa Univ.*)

Atomic force microscopy (AFM) has a unique capability to image nanoscale surface structures in liquid. This capability allows us to visualize structures and dynamics of proteins and cells in a biologically relevant solution. Owing to the recent development of AFM technologies, the variety, volume and complexity of the AFM data have been rapidly increasing. For example, the development of high-speed AFM enabled to capture many successive 2D images showing protein dynamics. The development of 3D-AFM enabled to produce 3D data representing local distribution of mobile water (i.e. hydration structure) and flexible molecular chains. The analysis of such higher-order AFM data is becoming increasingly difficult and hence requires advanced computational analysis method.

1SGP-7 細胞表面動態の高精度計測を目指した高速原子間力顕微鏡装置の開発
A High-speed atomic force microscope for detailed time series analysis of cell surface dynamics

Masahiro Shimizu[1,2], Chihiro Okamoto[3], Hirotatsu Imai[1], Shinji Watanabe[4], Toshio Ando[2,4], Noriyuki Kodera[2,4] ([1]*InFiniti, Kanazawa Univ.*, [2]*CREST, JST*, [3]*Dept. Sch. Math. & Phys., Kanazawa Univ.*, [4]*WPI-NanoLSI, Kanazawa Univ.*)

The devices limiting the scan-speed in the current high-speed atomic force microscopy (HS-AFM) are the Z-scanner and the amplitude detector of cantilever oscillation. Here, we developed a new Z-scanner with a resonant frequency over 1 MHz using a new holding method of piezo actuator. The speed of Z-scanner is now ~6-times improved. The narrowed Z-scan range was compensated by placing the new Z-scanner onto another wide range Z-scanner. In addition, the speed of the amplitude detector was improved by employing a new scheme. These improvements allow us to perform HS-AFM imaging with higher frame rate and sub-nanometer resolution in height even for microscale object, which would greatly contribute to the understanding of dynamic molecular phenomena happen on cell surface.

1SGP-8 高速原子間力顕微鏡 1 分子計測と分子シミュレーションのデータ同化による生体分子 4 次元構造解析
Four-dimensional biomolecular structure analysis with data assimilation of HS-AFM single molecule measurement and molecular simulation

Sotaro Fuchigami[1,2], Toru Niina[1], Shoji Takada[1,2] ([1]*Grad. Sch. of Science, Kyoto Univ.*, [2]*CREST, JST*)

High-speed atomic force microscopy (HS-AFM) is a powerful tool to observe structures and dynamics of single biomolecules at work in real time. However, its spatiotemporal resolution is not enough to reveal the molecular details of structural dynamics, and it is necessary to model its three-dimensional structure fitted into a HS-AFM image. In this presentation, we introduce our recent work on these problems. First, we have developed a flexible fitting method for a biomolecule to change its conformation so as to minimize the difference from a target AFM image. Next, we have developed a four-dimensional structure analysis method of single molecule measurement data by HS-AFM combined with coarse-grained molecular simulation using data assimilation with particle filter.

1SHP-1 Holistic Phenotyping of GPCR Signaling System by a Versatile Single-Platform Assay

Ikuo Masuho (*The Scripps Research Institute FL, Department of Neuroscience*)

Despite the eminent tractability of GPCRs as drug targets, there are substantial challenges in understanding the function of GPCRs and the mechanisms of drug action at these receptors. To address these challenges, we developed a single-platform assay for profiling GPCR activity on a nearly complete set of G protein substrates. Using this assay, we found 1) individual GPCRs exhibited characteristic "fingerprint-like" G protein-coupling profiles, 2) synthetic drugs impacted these fingerprints, and 3) certain genetic variants of receptors led to altered drug response. Thus, our approach will have a significant impact on phenotyping GPCR activity, realizing personalized medicine, and designing drugs to selectively engage therapeutically beneficial signaling pathways.

1SHP-2 GPCR の 1 分子拡散動態から複数の薬効を読み解く
Estimation of multiple drug effects on GPCR based on the single-molecule diffusion dynamics

Masataka Yanagawa (*Cellular Informatics Lab., Riken*)

G protein-coupled receptors (GPCRs) activate multiple signaling pathways through G proteins and arrestins upon ligand binding. Biased ligands with pathway-selective activity have been identified in various GPCRs, and have attracted much attention as drugs with lower side effects. However, it is difficult to evaluate the multiple activities of GPCR by a single measurement. Here, we show that drug effects on three signaling pathways downstream of S1PR1, a model GPCR, can be estimated simultaneously from the diffusion dynamics of receptor molecules in a single cell by single-molecule time-lapse imaging. We would like to discuss the future applicability of the present method in pharmacology and drug screening.

1SHP-3 Structure and conformational transitions of a neurotensin receptor 1 Gi1 protein complex

Hideaki Kato[1,3], Yan Zhang[2,3], Hongli Hu[3], Carl-Mikael Suomivuori[3], Francois Marie Ngako Kadji[4], Junken Aoki[4], Kaavya Krishna Kumar[3], Rasmus Fonseca[3], Daniel Hilger[3], Weijiao Huang[3], Naomi Latorraca[3], Asuka Inoue[4], Ron Dror[3], Brian Kobilka[3], Georgios Skiniotis[3] ([1]*The Univ. of Tokyo*, [2]*Zhejiang Univ.*, [3]*Stanford Univ.*, [4]*Tohoku Univ.*)

The neurotensin receptor 1 (NTSR1) is a GPCR involved in regulation of blood pressure, body temperature, and so on. NTSR1 couples to multiple G-protein subtypes, but the molecular details of G-protein activation remain unknown. Here we present 3-Å structures of the human NTSR1 in complex with the heterotrimeric Gi1 protein in two distinct conformations (C state and NC state). While the C-state complex is similar to other GPCR-Gi complexes, the G-protein and NTSR1 in the NC state exhibit unique features, suggesting that the structure may represent an intermediate. This structural information, complemented by MD simulations and functional studies, provides insights into the complex process of G-protein activation.

1SHP-4 NMR 法を用いた動的構造にもとづく GPCR のシグナル伝達機構の解明
Dynamics of G protein-coupled receptor related to various signaling revealed by NMR

Yutaka Kofuku[1], Yutaro Shiraishi[1], Mei Natsume[1], Junya Okude[1], Shunsuke Imai[1], Masahiro Maeda[2], Hideki Tsujishita[2], Takefumi Kuranaga[1], Masayuki Inoue[1], Kunio Nakata[3], Toshimi Mizukoshi[3], Takumi Ueda[1], Hideo Iwai[4], Ichiro Shimada[1] ([1]*Grad. Sch. Pharm. Sci., Univ. Tokyo*, [2]*Shionogi Co., Ltd.*, [3]*Ajinomoto Co., Inc.*, [4]*Univ. Helsinki*)

The C-terminal region of G-protein-coupled receptors (GPCRs), stimulated by agonist binding, is phosphorylated by GPCR kinases, and the phosphorylated GPCRs bind to arrestin, leading to the cellular responses. To understand the mechanism underlying the formation of the phosphorylated GPCR-arrestin complex, we performed NMR analyses of the phosphorylated β_2-adrenoceptor (β_2AR) and the phosphorylated β_2AR-β-arrestin 1 complex, in the lipid bilayers of nanodisc. Here we show that the phosphorylated C-terminal allosterically alters the conformation around the transemembrane helices 5 and 6. In addition, we found that the conformation induced by the phosphorylation is similar to that corresponding to the β-arrestin-bound state.

1SHP-5 Theoretical Prediction of Thermostabilizing Mutations for GPCR: Identification of Hot-Spot Residues to be Mutated Common in Class A GPCRs

Satoshi Yasuda[1,2,3], Yuta Kajiwara[4], Yuki Takamuku[1], Nanao Suzuki[1], Yosuke Toyoda[5], Kazushi Morimoto[5], Ryoji Suno[5], So Iwata[5], Takuya Kobayashi[5], Takeshi Murata[1,2], Masahiro Kinoshita[3] ([1]*Grad. Sch. Sci., Chiba Univ.*, [2]*MCRC*, [3]*IAE*, [4]*Grad. Sch. Ener. Sci., Kyoto Univ.*, [5]*Grad. Sch. Med., Kyoto Univ.*)

Structural determination of GPCRs has been hindered by their instability in detergent. Here we develop a theoretical prediction method for thermostabilizing mutations using our free-energy function which is focused on the translational entropy of nonpolar chains within the lipid bilayer as the entropic term and the protein intramolecular hydrogen bonding as the energetic term. Its high success rate is demonstrated for many GPCRs. Remarkably, we find that a mutation of $Ser^{3.39}$, a hot-spot residue, to lysine or arginine is commonly identified as the thermostabilizing mutation for many GPCRs of Class A in the inactive state. We have been successful in thermostabilizing several GPCRs, leading to the determination of new three-dimensional structures for two of them.

1SHP-6 (1Pos077) 理論計算による熱安定化ムスカリン M2 受容体の選択的アンタゴニスト AF-DX 384 結合型構造
(1Pos077) Structural insights into the subtype-selective antagonist binding to the M2 muscarinic receptor

Ryoji Suno[1], Sangbae Lee[2], Shoji Maeda[3], Satoshi Yasuda[4], Keitaro Yamashita[9], Kunio Hirata[5,6], Takeshi Murata[7], Masahiro Kinoshita[8], Masaki Yamamoto[5], Brian Kobilka[3], Nagarajan Vaidehi[2], So Iwata[8], Takuya Kobayashi[1] ([1]*Kansai Medi. Univ.*, [2]*City Hope Med. Ctr.*, [3]*Stanford Univ.*, [4]*Chiba Univ.*, [5]*RIKEN,SPring-8*, [6]*JST,PRESTO*, [7]*IAE, Kyoto Univ.*, [8]*Med,Kyoto Univ.*, [9]*Univ.Tokyo,Sci*)

Human muscarinic receptor, M2 is one of the five subtypes of muscarinic receptors belonging to the family of G protein-coupled receptors. We report high resolution structures of a thermostabilized mutant M2 receptor bound to a subtype selective antagonist AF-DX 384. Comparison of the crystal structures and pharmacological properties of the M2 receptor shows that the Arg in the S110R mutant mimics the stabilizing role of the sodium cation, that is known to allosterically stabilize inactive state(s) of class A GPCRs. Molecular Dynamics simulations reveal that tightening of the ligand-residue contacts in M2 receptor compared to M3 receptor leads to subtype selectivity of AF-DX 384.

2SBA-1 Chemical control of protein localization in the multimolecular cellular space

Shinya Tsukiji (*Grad. Sch. Eng., Nagoya Inst. Tech.*)

Cells regulate diverse signaling processes in a spatiotemporal manner by precisely controlling the localization of signaling proteins in the multimolecular, complex cellular environment. This presentation will focus on new chemical approaches to manipulate protein localization in living cells. We are developing small molecules that can relocate target proteins to specific intracellular locations, and applying them as tools to control various signaling molecules such as Ras, ERK, PI3K, and Rac. Our recent effort to create new synthetic organelles in living cells will also be presented.

2SBA-2 Secondary structure of DNA for liquid-liquid phase separation

Masahiro Mimura[1,2], Shunsuke Tomita[2], Ryoji Kurita[1,2], Kentaro Shiraki[1] ([1]*Pure and Appl. Sci., Univ. Tsukuba*, [2]*Biomed. Res. Inst., AIST.*)

Liquid-Liquid Phase Separation (LLPS) has been proposed as a mechanism for compartmentalization of intracellular components in crowded environment. It has recently been reported that LLPS controls fluid changes of chromatin structures in the presence of histones and single or double-stranded DNAs. Here, we study the role of secondary structure of single-stranded DNA (ssDNA) with histone H1 for forming liquid droplet by using model system. Histone H1 plays an important role for chromatin structures by binding to DNA in nucleosomes. Our results indicated that the unique structures of ssDNA promoted the formation of liquid droplet with histone H1. This suggests that various structures of genome DNA can control droplet formation within the chromatin structure.

2SBA-3 相分離生物学：相分離する LC ドメイン
Phasing Biology: Low-Complexity Domains Phase-Separate through Cross-β Interaction

Eiichiro Mori (*Dept. Future Basic Med., Sch. Med., Nara Med. Univ.*)

Protein segments of low sequence complexity/intrinsic disorder (LC-domains) are recognized to play roles in the basis of the membrane-less organelles through cross-β polymerization. These LC-domains are ubiquitously associated with regulatory proteins that control gene expression and RNA biogenesis, but they are also found in the central channel of nuclear pores, the nexus points of intermediate filament assembly, and the locations of action of other well-studied cellular proteins and pathways. Here in this talk, I would like to review the discoveries of the LC-domains and phase separation in the context of cell biology and biochemistry.

2SBA-4 分子夾雑系における光センサー蛋白質の動的挙動―揺らぎと反応ダイナミクス―
Fluctuation and reaction dynamics of a light sensor protein in crowding environment

Yusuke Nakasone, Hiroto Murakami, Masahide Terazima (*Grad. Sch. Sci., Kyoto Univ.*)

To understand the dynamic behavior of biomolecule reaction in crowding environment, we investigated the activity, reaction and fluctuation of a light sensor protein PAC under crowding condition. The PAC synthesizes a cAMP in a light-dependent manner. We found that the catalytic activity is suppressed dramatically under crowding condition. We also detected the reaction and the fluctuation in time domain by the transient grating method. Interestingly, the yield and rate of protein conformational change decrease under crowding condition, though the fluctuation is enhanced during the photoreaction which is similar to the case of the diluted condition. We will discuss the crowding effect as a reaction field of protein molecule based on these results.

2SBA-5 細胞様構造や細胞組織体の自己創生：分子夾雑系の活用
Self-emergence of primitive cell and cellular mini-organoids under crowding environment

Kenichi Yoshikawa (*Facul. Life Med. Sci., Doshisha Univ.*)

It will be reported that long double-stranded DNA (above the size of kbp) and F-actin are spontaneously localized within cell-sized droplets under crowding conditions with binary hydrophilic polymers. We also show that living cells exhibit specific localization/organization with microdroplets. A novel working hypothesis on the origin of life as well as on the self-organization of multi-cellular system is proposed.

2SCA-1 AMED-BINDS 事業におけるビッグデータ科学
Big Data Science at AMED-BINDS

Haruki Nakamura (*Institute for Protein Research*)

AMED-BINDS (Basis for supporting innovative drug discovery and life science research) program, started in April 2017 for five years. It covers structural biology, library screening, medicinal chemistry, genome analysis, and in-silico simulation studies, supporting early-stage drug discovery and basic life science. The program has been producing many excellent results both from the own researches of BINDS members and from supporting and assisting the other researchers, who submit their proposals to the BINDS members. Those results should be archived as Big Data in addition to publish papers, spreading them throughout the world. Here, we introduce the BINDS program and the essential issues of biological big data science, which are tried to be solved in the BINDS program.

2SCA-2 多層ニューラルネットワークを用いたタンパク質残基間コンタクトおよびタンパク質 - 基質相互作用の予測
Prediction of protein residue contacts and protein-ligand interactions with deep neural networks

Kentaro Tomii (*National Institute of Advanced Industrial Science and Technology (AIST)*)

We are in the era of abundant data such as nucleic and amino acid sequences, protein structures, and results of ligand binding assays. Using these data, we have developed methods for prediction of protein contacts and protein-ligand interactions based on deep neural networks. In both cases, we trained our neural networks in an end-to-end manner. We found that our methods outperformed existing ones, according to benchmarks. We expect that our methods would be able to accelerate drug discovery and life science research.

2SCA-3 Integrated approach of experimental data and computer modeling and simulation for understanding chromatin structure and dynamics

Hidetoshi Kono, Atsushi Matsumoto, Shun Sakuraba, Hisashi Ishida (*QST, Institute for Quantum Life Science (iQLS), MMS*)

X-ray crystallographic analysis and single molecule analysis of electron microscopy provide us precise images of biomolecules, but the information is basically static. X-ray or neutron scattering give us structural information of the molecules in solution, but they are low resolution. We integrate such information with aids of computer modeling and simulation to build the 3D structures in solution and investigate the dynamics of mono- or di-nucleosome which are fundamental structural units of chromatin. We show that histone tails, which are difficult to experimentally see because of their nature of being in intrinsically disordered, are a key to stabilize various conformations.

2SCA-4 電顕のインフォマティクス：2D 生画像データの収集と原子モデルのフィッティング
EM informatics: archiving raw 2D images and fitting atomic models into a map

Takeshi Kawabata, Genji Kurisu (*Inst.Prot.Res., Osaka Univ.*)

Cryo-electron microscopy (Cryo-EM) has recently emerged as a powerful technique to provide near-atomic resolution of maps and atomic models, which are stored in EMDB and PDB, respectively. Raw 2D images with huge file sizes are required to reconstruct and validate a 3D density map. We have established a mirror site of the EMPIAR database in Japan (https://empiar.pdbj.org), which stores public raw 2D EM images. We are also preparing to open the deposition site in PDBj. To bridge PDB and EMDB, we have developed the program “gmfit” to fit atomic models into a 3D density map. The calculation is fast with the help of Gaussian mixture model. The gmfit has improved to perform a partial fitting of an atomic model in a subspace of the 3D map, using masking function.

2SCA-5 Development of a deep-learning-based method to identify "good" regions of a cryo-EM grid

Tohru Terada[1,3], Yuichi Yokoyama[2], Kentaro Shimizu[1,3], Kouki Nishikawa[4], Yoshinori Fujiyoshi[4], Kazutoshi Tani[5] ([1]*III, Univ. Tokyo*, [2]*GSII, Univ. Tokyo*, [3]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [4]*Adv. Res. Inst., Tokyo Med. Dent. Univ.*, [5]*Grad. Sch. Med., Mie Univ.*)

Recent advances in cryo-electron microscopy (cryo-EM) have enabled protein structure determination at an atomic resolution. Cryo-EM specimens are prepared by rapidly freezing the protein solution on a metal grid coated with holey carbon film; this results in the formation of an ice film on each hole. The thickness of the ice film is a critical factor for high-resolution structure determination. Ice that is too thick degrades the contrast of the protein image, while ice that is too thin excludes the protein from the hole. Thus, trained researchers have to manually select "good" regions with an appropriate ice thickness. To alleviate this time-consuming burden, we have developed a deep learning-based method to identify such "good" regions from low-magnification EM images.

2SCA-6 クライオ電顕データ収集の効率化に資する凍結グリッド作成法やソフトウェア
Improvements in grid preparation method and software for facilitating cryoEM data collection

Keiichi Namba[1,2,3] ([1]*Graduate School of Frontier Biosciences, Osaka University*, [2]*RIKEN SPring-8 Center*, [3]*RIKEN Center for Biosystems Dynamics Research*)

Electron cryomicroscopy and single particle image analysis has become a powerful tool and now allows structural analysis to reach near-atomic resolution with a solution sample of only a few tens of micrograms. By using a new, fully automated cryoTEM, CRYO ARM 300, we developed with JEOL, we have been able to solve the structure of apoferritin at 1.53 Å resolution only from 840 images collected over one day. An extremely high coherence of the electron beam from its cold field emission gun made this highest resolution analysis possible. Now the bottlenecks of this technique are the reproducibility of cryoEM grid preparation with thin enough ice film for high-resolution analysis and the throughput of data collection. I will discuss some improvements in these two aspects.

2SDA-1 生物系のエナジェティクス
Energetics of biological system

Shoichi Toyabe (*Grad. Sch. Eng., Tohoku Univ.*)

Recent developments in nonequilibrium physics theories provide the methodology to measure and discuss the energetics of biological systems. Such theories have been applied to, for example, macromolecular stretching experiments, single molecular motor experiments, and recently cellular systems. The developments are revealing that the "information" is also an essential concept when we discuss the biological energetics. In this talk, I will briefly introduce basic concepts of biological energetics as an introduction for the following talks.

2SDA-2 歩行型モーター・キネシン 1 の非平衡エネルギー論
Nonequilibrium Energetics of a walking motor kinesin-1

Takayuki Ariga (*Graduate school of medicine, Yamaguchi University*)

Molecular motors are nonequilibrium open systems that convert chemical energy to mechanical work. We investigated the nonequilibrium energetics of a single molecule walking motor, kinesin-1. The sum of the dissipation measured from the violation of the fluctuation-response relation via the probe particle and the output power was only ~20% of the input free energy rate, indicating that large (~80%) internal dissipation exists [Ariga et al. Phys. Rev. Lett. 121, 218101. (2018)]. In this symposium, we discuss the origin of the dissipation by using a mathematical kinesin model and its apparent “low” energy efficiency by considering their intracellular working environment.

2SDA-3 FoF1-ATP 合成酵素の回転力はどのようにして発生しているのか? 構造生物・生物物理学的手法による解析
Structural and biophysical analyses of torque generation mechanism of F_oF_1-ATP synthase

Toshiharu Suzuki (*Lab for Chem and Life Sci, Inst of Innov Res, Tokyo Inst of Tech*)

F_oF_1-ATP synthase has dual-rotary motor architecture which is composed of ATPase-driven F_1-ATPase motor and H^+-flow-driven F_o motor, to achieve energy conversion between chemical potential of ATP and electrochemical potential of H^+ across membrane. Our recent X-ray crystallographic studies revealed catalytic intermediate states of binding/releasing of phosphate and ADP, and unveiled what structural rearrangements occur during the elementary steps. Additionally, high-resolution EM structure of *Bacillus* PS3 F_oF_1-ATP synthase (H Guo et al eLife2019) clarified an inevitable snapshot of torque generation site of F_o motor at atomic level. Together with results of biophysical analysis (N Soga et al PNAS2018), torque generation of the two motors will be discussed in this talk.

2SDA-4 Modeling of myosin V motor dynamics to understand high-speed AFM observations

Holger Flechsig (*WPI Nano Life Science Institute, Kanazawa University*)

In novel interactive high-speed AFM experiments an unprecedented stepping behavior of the myosin V motor along actin filaments has been observed, but the underlying mechanism is unknown. We have established a dynamical model which provides an explanation of those findings. I will explain the mechanism of single steps by the motor seen in the experiments, but also present a statistical analysis of motor operation from model simulations. Energetic aspects and a comparison with conventional motor stepping will be furthermore provided. Model results agree very well with experiments and a solid understanding of high-speed AFM observations could be obtained.

2SDA-5 Energetics and structural dynamics of a viral RNA polymerase ratcheting along DNA with fidelity control

Jin Yu (*Beijing Computational Science Research Center*)

RNA polymerase (RNAP) from bacteriophage T7 is a representative single-subunit viral RNAP. We studied this small transcription machine computationally as a model system to understand mechano-chemical coupling and fidelity control. Here we briefly present how T7 RNAP translocates along DNA in Brownian alike motions, and how correct nucleotide incorporation acts as a pawl for “selective ratcheting”. The structural dynamics and energetics features revealed from our atomistic molecular dynamics (MD) simulations and related analyses on the single-subunit T7 RNAP thus provided detailed and quantitative characterizations on the Brownian-ratchet working scenario of a prototypical transcription machine with sophisticated nucleotide selectivity for fidelity control.

2SDA-6 Error-speed correlations in biopolymer synthesis

Simone Pigolotti (*Okinawa Institute of Science and Technology Graduate University*)

Biological systems are able to replicate information with outstanding accuracy. In biochemical pathways such as those leading to DNA duplication or protein translations, different monomers can be distinguished because of equilibrium free-energy differences or via non-equilibrium mechanisms. In the first part of my talk I will show how, in simple copying reactions, these two discrimination modes are mutually exclusive and lead to opposite tradeoffs between error, dissipation and reaction velocity. In multi-step reactions, such as in kinetic proofreading, these different modes can be combined to improve overall accuracy. In the second part of my talk, I will present recent results about fluctuations in accuracy and speed of these reactions.

2SDA-7 (2Pos121) Dynamic energy landscape of a linear motor chitinase from single-particle tracking trajectories

Kei-ichi Okazaki, Akihiko Nakamura, Ryota Iino (*Institute for Molecular Science*)

Single-molecule experiments have been used to investigate mechanism of biomolecular motors by visualizing their motions. In this study, we estimate an underlying diffusion model from observed single-molecule trajectories. To deal with nonequilibrium trajectories driven by chemical energy consumed by biomolecular motors, we develop a novel framework based on hidden Markov model, which considers switching among multiple energy surfaces depending on chemical states of the motors. The method is applied to single-molecule trajectories of processive chitinase, a linear motor that moves with hydrolysis energy of single chitin chain. The dynamic free energy landscape underlying the burnt-bridge Brownian ratchet mechanism of chitinase is revealed.

2SEA-1 非結晶生体試料のシンクロトロン放射光 X 線回折実験に関する最近の進歩
Recent progress in synchrotron radiation X-ray diffraction studies for non-crystalline biological specimens

Hiroyuki Iwamoto (*SPring-8, JASRI*)

The X-ray diffraction technique has a potential to image non-crystalline biological specimens at a near-atomic resolution. To do this, both the amplitude and the phase of the diffraction pattern are required, but the phase is lost when the pattern is recorded by a detector. This “phase problem” has been the largest problem for the X-ray diffraction technique. However, this situation has changed by the recent developments of algorithms to recover the once-lost phase information and restore the structure of the sample in the real-space. This technique is called “lenseless imaging”. In this talk the recent development of “lenseless imaging” is reviewed and the possibility of its application to the imaging of non-crystalline biological specimens is discussed.

2SEA-2 X 線繊維回折法によって明らかとなった秒単位の微小管構造変化
Dynamic changes of tubulin dimer configurations on a scale of sub-second revealed by high flux X-ray fiber diffraction

Shinji Kamimura[1], Hiroshi Imai[2], Toshiki Yagi[3], Hiroyuki Iwamoto[4] ([1]*Dept. Biol. Sci., Chuo Univ.*, [2]*Grad. Sch. Sci., Osaka Univ.*, [3]*Dept. Life Sci., Prefect. Univ. Hiroshima*, [4]*SPring-8, JASRI*)

Microtubules are assembled from tubulin dimers that are arranged in a semi-crystal lattice with high regularity. Dynamics of tubulin dimer within microtubules is thus expected to be directly affecting the stability of microtubules, however, it has been difficult to quantitatively describe such properties. By analyzing the time course of pattern changes in X-ray fiber diffractions from aligned microtubules with a high-flux synchrotron beam of BM40XL (SPring-8), we found microtubules showed rapid elongation in the axial tubulin repeat and the concomitant increase of structural flexibility with a time constant of about 0.2 s after applying paclitaxel, microtubule-stabilizer. Contrasting effects were found for laulimalide, another type of microtubules stabilizer.

2SEA-3 Diffracted X-ray Tracking for protein dynamics

Hiroshi Sekiguchi (*Cent. Synchrotron Rad. Res., JASRI/SPring-8*)

At synchrotron radiation facilities, we can utilize bright light with a wide energy range of X-rays. In the field of life sciences, many achievements have been made particularly in protein crystal structure analysis, and dynamics of proteins, such as structural changes, association, and detachment of ligand molecules in solution can be obtained by time-resolved diffraction or scattering measurement. In this presentation, we will introduce an application of the diffracted X-ray tracking and blinking method that utilizing the feature of synchrotron radiation. In these techniques, gold nanocrystal is used as motion probe, and the trajectory of its position or intensity of diffracted spot are investigated to elucidate the motion of the protein (Sci. Rep 2014, 2018).

2SEA-4 Dynamic changes in cardiac myosin head regulation during hyperglycemic events in insulin resistant rats

James T. Pearson[1,2], Naoto Yagi[3], Mikiyasu Shirai[1], Mark Waddingham[1], Hirotsugu Tsuchimochi[1], Takashi Sonobe[1], Vijayakumar Sukumaran[1] ([1]*National Cerebral and Cardiovascular Center*, [2]*Monash University, Department of Physiology*, [3]*JASRI*)

In cardiomyocytes, myosin head displacement from proximity to actin filaments contributes to impaired muscle fibre relaxation and diastolic dysfunction in insulin resistant (Goto-Kakizaki) rats. Increased reactive oxygen species (ROS) generation potentially contributes to this dysfunction. Utilising synchrotron X-ray scattering we examined the effect of acute oral glucose loading on in situ cross-bridge cycling in the left ventricle of GK and Wistar rats. We found that myosin head displacement due to a transient increase in blood glucose occurred in non-diabetic Wistar rat hearts (subendocardium only). On the other hand, displacement was exacerbated in GK rats throughout the ventricle wall and was sustained for 2h. The role of ROS generation is being examined.

2SEA-5 (2Pos269) G1 期酵母細胞核内における核酸分布の XFELX 線回折イメージング
(2Pos269) Distribution of nucleic acids in yeast nucleus of G1 phase visualized by X-ray diffraction imaging using X-ray free electron laser

Masayoshi Nakasako[1,2], Takahiro Yamamoto[1,2], Amane Kobayashi[1,2], Mao Oide[1,2], Koji Okajima[1,2], Yuki Takayama[1,2,3], Tomotaka Oroguchi[1,2], Masaki Yamamoto[2] ([1]*Keio University*, [2]*RIKEN*, [3]*University of Hyogo Prefecture*)

Distribution of nucleic acids in yeast nuclei was visualized by X-ray diffraction imaging using X-ray free electron laser. We obtained high quality diffraction patterns of frozen-hydrated yeast nuclei at the G1 phase. From the diffraction patterns, we first found mass and surface fractal properties with respect to the packing of substructural elements. The projection electron density maps retrieved form diffraction patterns indicated non-uniform distribution of nucleic acids including related proteins. From the maps, we speculated a model with a U shape to explain most of projection maps observed.

2SFA-1 記憶痕跡サブ・アンサンブルの協奏的活動によるエピソード記憶の脳内表現
Orchestrated ensemble activities constitute a hippocampal memory engram

Noriaki Ohkawa[1,2] ([1]*Univ of Toyama Grad Sch of Med and Pharm Sci*, [2]*PRESTO, JST*)

The brain stores memories through a set of neurons, termed engram cells. It is unclear how engram cells are organized to constitute a corresponding memory trace. We established a unique imaging system that combines Ca2+ imaging and engram identification to extract the characteristics of engram activity. We detected several sub-ensembles composed of neurons collectively activated during learning. Some sub-ensembles of engram, but not non-engram, preferentially reappear during post-learning sleep, and these replayed sub-ensembles are more likely to be reactivated during retrieval. These results indicate that engram sub-ensembles represent distinct pieces of information, which are then orchestrated to constitute an entire memory.

2SFA-2 X線を用いた神経機能の遠隔無線操作
Remote and wireless control of neuronal function using X-ray

Takayuki Yamashita[1] ([1]*Dept. Neurosci. II, RIEM, Nagoya Univ.*, [2]*PRESTO, JST*)

Scintillators exhibit visible luminescence called scintillation when excited by X-ray. We found that a certain yellow-emitting scintillator, which is biocompatible, can efficiently activate red-shifted opsins (light-sensitive ion channels) upon X-irradiation. Taking advantage of the excellent capability of X-ray to penetrate biological tissues, we successfully activated and inhibited deep brain cells of freely moving mice to wirelessly drive related behaviors under X-irradiation. Our data highlight the potential of X-ray-induced scintillation to be used for distant control of cellular functions in the brain and other organ systems of living animals including humans.

2SFA-3 バッテリレス超小型光刺激デバイス
Batteryless ultra-small implantable optical stimulator

Takashi Tokuda[1], Thanet Pakpuwadon[2], Nattakarn Wuthibenjaphonchai[2], Makito Haruta[2], Kiyotaka Sasagawa[2], Jun Ohta[2] ([1]*FIRST, Tokyo Tech*, [2]*Mater. Sci., NAIST*)

An ultra-small, electronics-based implantable optical stimulator is proposed and demonstrated. The size of the device is as small as 1mm3 and the weight of the device is 2.3mg. The device is operated by IR light that can be deliverd to the device through skin and tissues. The device has multiple photovoltaic cells and charge the energy generated from IR light. After a sufficient energy is accumulated, the device perform an optical stimulation. This device can be operated with reasonable irradiation of the IR light, differing from nanomaterial-based upconversion schemes.

2SFA-4 長波長レーザーによる超深部イメージングシステムの開発
Development of deep-tissue imaging system based on a long-wavelength laser

Yutaka Nomura (*Institute for Molecular Science*)

The imaging depth in biological tissues is dependent on the wavelength of the excitation laser beam and dominated by the absorption and the scattering within the tissues. Since the scattering effect generally becomes smaller as the wavelength becomes longer, the penetration depth can be significantly increased by using a long-wavelength radiation as the excitation light source for multi-photon microscope. Here, I present an ultrafast laser operating at 1.8 μm based on a thulium-doped fiber laser and its application to a multiphoton microscope. The developed system will be useful for sending the light to unprecedented depth in biological tissues and hopefully open up new horizons in biological science and optogenetics.

2SFA-5 光操作型アデノウイルスベクターの開発と応用
Generation of photoactivatable adenovirus vector for spatiotemporally controllable gene therapy

Kazuo Takayama[1,2,3] ([1]*Osaka University*, [2]*NIBIOHN*, [3]*PRESTO*)

Gene therapy is expected to be utilized for the treatment of various diseases. However, the spatiotemporal resolution of current gene therapy technology is not high enough. In this study, we generated a new technology for spatiotemporally controllable gene therapy. We introduced optogenetic and CRISPR-Cas9 techniques into a recombinant adenovirus (Ad) vector to generate an illumination-dependent spatiotemporally controllable gene regulation system (designated the Opt/Cas-Ad system). With the Opt/Cas-Ad system, highly selective tumor treatment could be performed by illuminating the tumor. We believe that our Opt/Cas-Ad system can enhance both the safety and effectiveness of gene therapy.

2SFA-6 (2Pos198) 微生物型ロドプシンに基づく光遺伝学ツールの探索と開発
(2Pos198) Exploration and development of microbial rhodopsin-based optogenetic tools

Keiichi Kojima, Yuki Sudo (*Grad. Sch. of Med. Dent. Pharm. Sci., Okayama Univ.*)

Microbial rhodopsins are photoreceptive membrane proteins containing retinal as a chromophore. Genomic advances have revealed that they are widely distributed in all three biological domains with a wide variety of biological functions (e.g. ion transporters and light sensors). Recently, they serve as fundamental tools of optogenetics, a technology to regulate biological phenomena with light. Towards development of optogenetic tools, we are investigating them with bottom-up strategies as follows; (i) we explored novel rhodopsins from nature and characterized their functions and properties. (ii) we rationally modified their properties. (iii) we applied them to optogenetic tools. In this symposium, we will discuss our recent progress and future prospects.

2SFA-7 「総力戦」としての光操作技術
Optical control as a fusion of neuroscience, medicine, engineering, and biophysics

Hisao Tsukamoto[1,2] ([1]*Institute for Molecular Science*, [2]*PRESTO, JST*)

Optical control is a major contributor of recent advances in biology. In particular, optogenetics introducing photoreceptive proteins such as channelrhodopsin into specific neural cells has revolutionized neuroscience, and the concept has also been applied to other fields such as biomedical research. To elucidate or manipulate biological functions by optical control, you need to fuse various research fields. Genomics, biophysics, and synthetic chemistry provide light-activatable molecules day by day, and engineering develops state-of-art devices delivering light more easily and more precisely. Here I will give an overview of such efforts based on the presentations in this symposium, and also introduce new GPCR-based tools for optical control.

2SGA-1 生体内の電子伝達金属補因子「鉄硫黄クラスター」の生合成機構
Molecular mechanism of the biosynthesis of the iron-sulfur clusters involved in the electron transport in vivo

Kei Wada (*Dept. of Medical Sciences, Univ. of Miyazaki*)

Iron-sulfur (Fe-S) clusters are found in all domains of life and have evolved to play roles in many biological processes, both as electron transfer and as more complex redox catalysts. In Fe-S proteins, iron atoms are bridged by inorganic sulfur atoms in the form of [4Fe-4S] and [2Fe-2S] clusters, which are further ligated to the polypeptides primarily via the thiolate side chains of cysteines. The redox potential of Fe-S cluster is controlled by it binding environment (-700 mV to +400 mv). SUF system is most widely distributed pathway for Fe-S cluster, and the SufBCD complex is a core component that is responsible for de novo cluster biogenesis. In this study, the crystal structure of the complex provided insights into the molecular mechanism of Fe-S cluster assembly.

2SGA-2 高エネルギー硫酸ヌクレオチド分子を利用した生体内代謝：硫酸転移酵素の多様な生理機能の解明
Metabolism of key endogenous molecules mediated by sulfotransferases with a hi-energy sulfonucleotide, PAPS

Katsuhisa Kurogi[1], Takamasa Teramoto[2], Yoshimitsu Kakuta[2], Ming-Cheh Liu[3], Masahito Suiko[1], Yoichi Sakakibara[1] ([1]*Dept. Biochem. Appl. Biosci., Fac. Agric., Univ. Miyazaki*, [2]*Dept. Biosci. Biotechnol., Grad. Sch. Agric., Kyushu Univ.*, [3]*Dept. Pharmacol., Coll. Pharm., Univ. Toledo*)

Sulfonation is an important and well-known pathway involved in the metabolism of drugs and key endogenous molecules including steroid/thyroid hormones and neurotransmitters. Sulfotransferases, responsible for the sulfonation reaction, transfer a sulfonate group from a hi-energy sulfonucleotide, 3'-phosphoadenosine 5'-phosphosulfate (PAPS), to a hydroxyl or an amino group of a substrate molecule. PAPS is generated from 2 ATP and a sulfate ion in cytosol and sulfation reaction bi-product, 3'-phosphoadenosine 5'-phosphate (PAP) is then metabolized into 5'-AMP. My topics will be focused on the reaction mechanism of sulfonate transfer and the physiological significance of sulfonate transfer from a hi-energy sulfonucleotide to target molecules.

2SGA-3 発熱植物の生体エネルギー論：シアン耐性呼吸からミトコンドリアの構造まで
Towards understanding the roles of mitochondrial energy bypasses in heat-producing plants

Yasuko Ito-Inaba (*Agric, Univ. Miyazaki*)

Heat production in flowers or cones has been observed in several primitive seed plants, and so far, around 80 species are recognized as thermogenic species. In these species, the most commonly proposed role for thermogenesis is to promote the emission of volatiles that may serve to attract pollinators. The role of mitochondrial energy bypasses, especially alternative oxidase (AOX) pathway, in plant thermogenesis has been studied in several thermogenic aroids, sacred lotus, and recently, in cycad species, *Cycas revoluta* (Ito-Inaba et al., *Plant Physiol*., 2019). In the symposium, combining previous reports with our recent findings, I would like to focus on how thermogenic respiratory processes, including AOX respiration, drive the increase of flowers/cones temperature.

2SGA-4 The role of Fhod family formin proteins in mouse heart

Fumiyuki Sanematsu[1], Hideki Sumimoto[2], Ryu Takeya[1] ([1]*Dept. of Pharmacol., Fac. of Med., Univ. of Miyazaki*, [2]*Dept. of Biochem., Kyushu Univ. Grad. Sch. of Med. Sci.*)

Sarcomere, a contractile apparatus composed of actin and myosin filament, has the ability to cause contraction of muscle cells by converting chemical energy into the mechanical work. Organization and maintenance of straight actin filaments are required for efficient sarcomere contraction mediated by myosin ATPases. Fhod3, an actin-organizing formin protein, is a key regulator for this process in cardiomyocyte. Fhod3 null mice exhibit defects of cardiac development and the contractile function. In contract, depletion of Fhod1, a protein closely-related to Fhod3, does not lead to any defects in the heart. In this symposium, we will present the latest findings of the role of Fhod proteins in mouse heart and discuss the possible mechanisms involved.

2SGA-5 In-cell ^{19}F NMR: Chemical method for investigating nucleic acid structure in living cells

Takumi Ishizuka, Yan Xu (*Fac. Med., Univ. of Miyazaki*)

The various conformations of DNA and RNA have important biological roles. Investigation of the structures DNA and RNA is therefore essential to an understanding of the functions of nucleic acids. In the present studies, we developed a ^{19}F NMR spectroscopy method to investigate the telomeric RNA and DNA G-quadruplex structures *in vitro* and in living cells. By employing the ^{19}F NMR, we confirmed that telomere RNA forms the higher-order G-quadruplex in *Xenopus laevis* oocytes. We further demonstrated that the telomere DNA sequence can form two hybrid-type G-quadruplex structure in living human cells (HeLa cells). The results indicated that ^{19}F NMR is a useful method for the investigation of nucleic acid structures *in vitro* and in living cells.

2SGA-6 微生物電池における c 型シトクロムを介した細胞外電子伝達
Extracellular electron transfer via c-type cytochromes in microbial fuel cells

Kengo Inoue (*Department of Biochemistry and Applied Biosciences, Faculty of Agriculture*)

MFCs are devices which can convert chemical energy of organic compounds to electric energy by electrochemically active microorganisms. Geobacter sulfurreducens produces current at higher current density than any other microorganisms. This microorganis has over 100 c-type cytochromes, which play important roles in extracellular electron transfer. Among the cytochromes, OmcZ is essential for optimal current production. OmcZ undergoes complex procedure, such as translocation to periplasmic site, heme maturation, secretion across outer membrane, and cleavage by subtilisin-like protease. The process increase insolubility of OmcZ, which is thought to be important for affinity to electrode with hydrophobic properties.

2SHA-1 生物学的ネットワークの確率論的変動
Stochastic usage of biological network

Takuma Shiraki (*Kindai Univ. BOST*)

Development of a research filed of molecular biology from genetics revealed a vast of molecules involved in various biological responses. Molecules, usually proteins, form biological network to have a certain function, however, we misunderstand that a MOLECULE has a function. Rather, we should interpret it as a NETWORK has a function. In this presentation, I will explain how each cell uses distinct biological network, one-by-one, resulting in producing cell-to-cell variation. In addition, this variability enables cells to respond to various kind of stimuli. We will introduce a novel analytical method utilizing this stochastic usage of network to evaluate the contribution of molecules involved in a cellular response that you are interested in.

2SHA-2 Autoencoder-based analyses of dynamic allostery on proteins by regulator binding

Yuko Tsuchiya[1], Kei Taneishi[2], Yasushige Yonezawa[3] ([1]*AIRC, AIST*, [2]*RIKEN*, [3]*KINDAI University*)

Analyses of subtle changes in side-chain dynamic motion, and not large rigid conformational changes, are essential to understand the regulation of protein functions. We propose an autoencoder-based method that can detect dynamic allostery, based on the comparison of protein fluctuations, using distance matrices obtained from the molecular dynamics simulations in regulator-bound and -unbound forms. The method detected that the changes in dynamics by regulator binding led to the reorganization of correlative fluctuations among residue pairs. Other correlative motions were also found as a result of dynamic perturbation. Our method would be usefully applied to the signal transduction and mutagenesis systems that are involved in protein functions and severe diseases.

2SHA-3 大規模量子分子動力学シミュレーションを用いた光受容タンパク質におけるプロトン移動反応の解明
Clarification of proton transfer reactions in photoreceptive proteins using large-scale quantum molecular dynamics simulations

Junichi Ono[1], Chika Okada[2], Yoshifumi Nishimura[1], Hiromi Nakai[1,2,3] ([1]*WISE, Waseda Univ.*, [2]*Grad. Sch. of Adv. Sci. & Eng., Waseda Univ.*, [3]*ESICB, Kyoto Univ.*)

Photoreceptive proteins transduce light into energy or signal, and the proton transfer reactions play a vital role in achieving this function. However, these reactions have not been directly observed due to the limit of the spatio-temporal resolution in experiments. Thus, the microscopic mechanisms remain unclear. In our group, the divide-and-conquer density-functional tight-binding (DC-DFTB) method has been developed for large-scale quantum molecular dynamics simulations, and applied to chemical reactions in condensed phases in conjunction with metadynamics, one of the enhanced sampling methods. Here, our recent applications to proton transfers in photoactive yellow protein and bacteriorhodopsin (in total ~16,000 and ~50,000 atoms, respectively) will be discussed.

2SHA-4 カルシウムシグナル伝達蛋白質 Calmodulin と結合ドメインの構造変化と相互作用
Conformational changes and interactions of calcium ion signal transfer protein Calmodulin and Calmodulin-binding domain

Hiromitsu Shimoyama (*Kitasato-Univ.*)

Calmodulin (CaM) is a major calcium ion signal transfer protein in many cellular processes. CaM undergoes large conformational change upon calcium ion binding. The conformational change is necessary to form complex with its target protein's binding domain (CaMBD; about 20-residue long peptide). However, the process of conformational change and complex forming are still not well understood. Recently I analyzed various structures of CaM and clarified interactions driving the conformational change. In this work, mechanism of CaM's conformational change and binding to CaMBD is studied by multiscale molecular dynamics and rigid-body docking.

2SHA-5 タンパク質中の不均一なエネルギー流と機能に関する理論的研究
Theoretical study on non-uniform energy flow and protein function

Genki Kubota, Olivier Laprevote, Takahisa Yamato (*Nagoya University*)

Within thermally fluctuating protein molecules under physiological conditions, tightly packed amino acid residues interact with each other through heat and energy exchanges. We have developed a computer program, CURP (CURrent calculations for Proteins), which is suitable for analyzing non-uniform flow of heat and energy in proteins (http://www.comp-biophys.com/yamato-lab/curp.html). We illustrate and characterize non-uniform pattern of heat flow in proteins with the energy exchange network (EEN) model based on “local heat conductivity” between each residue pair. Visual representations of EEN diagrams demonstrated characteristic features of “hidden dynamic allostery” in PDZ domain and allosteric transition in the oxygen sensor domain of FixL.

2SHA-6 (2Pos057) ダイナミン GTP アーゼはアクチン線維の束化と分散を機械的に制御する
(2Pos057) Dynamin GTPase mechanically regulates bundling and unbundling of actin filaments

Kohji Takei[1], La The Mon[1], Tadashi Abe[1], Tetsuya Takeda[1], Ikuko Fujiwara[2], Akihiro Narita[3] ([1]*Grad.Sch. Med.Dent. Pharm.Sci., Okayama Univ.*, [2]*Dept. Biol.Facul.Sci., Osaka City Univ.*, [3]*Struct. Biol. Res. Ctr and Divi. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

Actin cytoskeleton is dynamically remodeled in variety of cellular events. We previously found that dynamin bundles F-actin (Yamada et al. J Neurosci. 2013; Int. J. Oncol. 2016). However, the molecular mechanisms are poorly understood. Here we show that dynamin mechanically bundles F-actin by polymerizing into helix around several actin filaments. Dynamin GTPase activity was drastically enhanced by the presence of F-actin. Addition of GTP to F-actin bundles formed by dynamin lead to rapid depolymerization of dynamin and unbundling of actin filaments. Furthermore, we found that ionic strength affects both dynamin polymerization and actin bundle formation. Thus, bundling and unbundling of F-actin is strongly linked to dynamin’s self-polymerization and GTP hydrolysis.

2SHA-7 Analysis of Effect of Mutation on the Response for Membrane Depolarization in the Voltage-Gated Potassium Channel Kv1.2

Hiroko X. Kondo[1], Norio Yoshida[2], Gen Masumoto[3], Matsuyuki Shirota[4,5,6], Yu Takano[7], Kengo Kinoshita[5,6] ([1]*Fac. Eng., Kitami Inst. Tech.*, [2]*Grad. Sch. Sci., Kyushu Univ*, [3]*RIKEN ISC*, [4]*Grad. Sch. Med., Tohoku Univ.*, [5]*GSIS, Tohoku Univ.*, [6]*ToMMo, Tohoku Univ.*, [7]*Grad. Sch. Info. Sci., Hiroshima City Univ.*)

Voltage-gated potassium (Kv) channels conduct potassium ions selectively in response to membrane depolarization and regulate the amplitude and duration of action potentials. Ion permeation through the pore domain is dynamically regulated by several types of gating mechanisms, and a mutation alters the ion conduction. To clarify the effect of mutations on the response for the change of membrane potential, we performed molecular dynamics simulations of the wild-type and mutant proteins of the Kv1.2-2.1 chimera channel and carried out the structural analysis by using the statistical methods. To evaluate a correlation between the conformation and ion occupancy state, we also analyzed the probability distributions of potassium ions by using the 3D-RISM theory.

2SBP-1 トランスオミクスに資する次世代メタボローム分析技術の開発
Development of next generation metabolome analytical technologies for trans-omics

Takeshi Bamba (*Med. Inst. Bioreg., Kyushu Univ.*)

Many researchers have been working on the development of various metabolomics technologies for more than 10 years. However, there remains room for improvement in the coverage of metabolites, quantitative performance, reproducibility, differences between experimental errors in different batches, etc. Our group is working on developing technologies of metabolome analysis that could contribute to trans-omics research. In particular, we have been actively involved in the progress in various technological developments such as production of stable isotope-labeled standards and probes for quantitative analysis, quantitative wide-targeted metabolome analytical systems, and individual quantitative analytical systems.

2SBP-2 Novel sequencing methods to read the epigenome

Takashi Ito (*Kyushu Univ. Grad. Sch. Med. Sci.*)

The genome exerts its functions through interactions with a variety of chromatin proteins. Various epigenetic modifications affect these interactions directly or indirectly, thereby modulating genome functions. Hence comprehensive identification of genomic sites occupied by proteins and those bearing epigenetic modifications is vital to understand the epigenome, which is, by its nature, composed of multiple layers and affected by cellular metabolic state. In this presentation, I will introduce our efforts to develop novel sequencing methods to read the epigenome, including PBAT-seq for DNA methylation, DMS-seq for protein-DNA interactions, and others.

2SBP-3 Sulfur Metabolism Rewiring in NRF2-Addicted Cancer Cells

Hozumi Motohashi (*Institute of Development, Aging and Cancer, Tohoku University*)

KEAP1-NRF2 system is a sulfur-employing defense mechanism. NRF2 exerts potent anti-oxidant function and anti-inflammatory function. Although previous studies also reported that NRF2 promotes mitochondrial activity, its precise mechanism has not well understood. Our recent study on sulfur metabolism clarified NRF2 role in the regulation of mitochondrial energy metabolism. We also found that NRF2-addicted cancers, in which NRF2 is aberrantly activated and drives malignant progression of cancer, exploit completely different sulfur metabolism. This rewiring of sulfur metabolism is coupled with unique glutamine metabolism and regarded as metabolic adaptation of NRF2-addicted cancer cells to acquire potent cell survival capacities.

2SBP-4 条件付き独立性を用いたネットワーク構造推定
Network structure inference by conditional independence

Shinsuke Uda (*Kyushu university*)

Conditional mutual information is a useful measure for detecting the association between variables that are affected by other variables. Though permutation tests are used to check whether the conditional mutual information is zero to indicate mutual independence, permutations are difficult to perform because the other variables in a dataset may be associated with the variables in question. This study aims to propose a computational method for approximating conditional mutual information based on the distribution of residuals in regression models of the associations in question. The proposed method can implement the permutation tests for statistical significance by translating the problem of measuring conditional independence into the problem of estimating simple independence.

2SBP-5 一細胞遺伝子発現プロファイルからの細胞集団および細胞間コミュニケーションの動態モデリング
Modeling dynamics of cell population and cell-to-cell communication from single-cell gene expression profiles

Teppei Shimamura (*Nagoya University*)

Dynamics of cell population and cell-to-cell communication have critical roles in deciphering diverse organismal processes of multicellular systems including metabolic adaptation. Recent high-dimensional single-cell technologies have provided the ability to investigate expression patterns at the resolution of a single cell and characterize cell populations, cell types, and cell states. In this presentation, we present simple and efficient statistical frameworks to perform clustering and cell population tracking for time-series flow and mass cytometry data. We also provide a novel Bayesian framework using tidy representation for non-negative tensor factorization, to discover cell-to-cell communications and their associations with environmental factors.

2SBP-6 オミクスを数理モデルでつなぐ
Analyze Omics data using kinetic model

Mariko Okada[1] ([1]*Institute for Protein Research, Osaka University*, [2]*RIKEN IMS*)

Gene regulation is not only limited to transcriptional regulation, but is also maintained via crosstalk between other omic layers, including protein, DNA, and metabolic regulation. Thus, gene regulatory networks inheritably constitute a large number of omics-to-omics interactions. Our laboratory has been working for mathematical modeling of signaling network, of which system is controlled by temporal interactions of a relatively small number of proteins and enzymes. We now try to understand how those molecules control the expression of a large number of target genes. Our analysis combined with kinetics model and omics analysis indicated that the magnitude of cooperativity in enhancer regulation may function for quantitative control of target gene expression.

2SCP-1 アルツハイマー病の解明のためにシンギュラリティ生物学ができること
What singularity biology can do for understanding Alzheimer's disease

Hiroko Bannai[1,2], Michio Hiroshima[3], Yoshiyuki Soeda[4], Akihiko Takashima[4] ([1]*Keio Univ. Sch. Med.*, [2]*JST ERATO*, [3]*RIKEN BDR*, [4]*Gakushuin Univ, Faculty. Sci.*)

Alzheimer's disease (AD) starts from the accumulation of toxic Tau in a few neurons in a small region of the brainstem, and results in massive neuronal death in 50% of cerebral cortical neurons. Therefore, Tau accumulation process in AD is considered to be a singularity phenomenon that is originated from small numbers of cells. How are the first cells to propagate toxic Tau oligomers generated? When and how Tau oligomers propagate throughout the brain? Here, we will discuss how we can answer these biological questions that leads to understanding of AD. We will investigate the molecular mechanism of Tau propagation using single molecule imaging technique and inhibitors for Tau aggregation, and will develop novel probe to visualize Tau oligomers.

2SCP-2 Delineation of the activation trajectory of autoreactive T cells

Taku Okazaki, Hikari Okamura, Il-mi Okazaki, Kenji Shimizu, Takumi Maruhashi, Daisuke Sugiura (*Div Imm Reg, Inst Adv Med Sci, Tokushima Univ*)

The recent success of tumor immunotherapy targeting immuno-inhibitory co-receptors, PD-1 and CTLA-4 and the concomitant development of immune-related adverse events highlight the importance of inhibitory co-receptors in the regulation of cancer immunity as well as autoimmunity. However, how T cells specific to tumor-associated antigens or self-antigens are activated to eradicate tumors or damage self-tissues are not well understood. We have been trying to elucidate how inhibitory co-receptors regulate the activation of these T cells to attenuate cancer immunity or avoid autoimmunity by delineating the activation trajectory of T cells in the presence or absence of inhibitory co-receptors.

2SCP-3 全脳イメージングシステム FAST を用いたアンバイアスで仮説に依らない脳内シンギュラリティの検出
Unbiased and hypothesis-free approach to detect singularity in the brain using whole-brain imaging system FAST

Hitoshi Hashimoto[1,2,3,4], Takanobu Nakazawa[1,5], Kaoru Seiriki[1,6], Atsushi Kasai[1] ([1]*Lab. of Mol. Neuropharmacol., Grad. Sch. of Pharmaceut. Sci., Osaka Univ.*, [2]*Center for Child Mental Dev, United Grad. Sch. of Child Dev., Osaka Univ.*, [3]*Div. of Biosci., Inst. for Datability Sci., Osaka Univ.*, [4]*Transdimensional Life Imaging Div., Inst. for Open and Transdisciplinary Res. Initiatives, Osaka Univ.*, [5]*Dep. of Pharmacol., Grad. Sch. of Dentistry, Osaka Univ.*, [6]*Institute for Transdisciplinary Grad. Degree Programs, Osaka Univ.*)

Neurons of the mammalian central nervous system are highly diverse in their development, structure and function. In this study, we aim to investigate the molecular pathophysiology of brain disorders by developing automated high-speed imaging system (block-FAce Serial microscopy Tomography, FAST) and establishing mouse disease models and iPS cell-derived neurons from patients. For this aim, we have particular interest in identifying "brain singularity biology (cells, circuits, phenomena, mechanisms)" through unbiased and hypothesis-free approaches. In this symposium, we will introduce our recent progress and wish to discuss the directions of future research that would contribute to a better understanding of the brain systems and better treatments for brain disorders.

2SCP-4 顕微鏡ライブイメージングと 1 細胞 RNA-seq を組み合わせた自動化システムの開発とシンギュラリティ生物学への応用
An automated system for combining single-cell RNA-seq with live cell imaging and its applications for Singularity Biology

Taisaku Ogawa[1], Katsuyuki Shiroguchi[1,2] ([1]*RIKEN BDR*, [2]*RIKEN IMS*)

Most cellular dynamics can be visualized by live cell imaging. To understand the molecular mechanism of them, it is required to investigate the internal state of the individual cells by, e.g., RNA sequencing for imaged cells. However, conventional sequencing platforms require flow cytometers or droplet microfluidics to isolate single cells. Therefore, it is difficult to perform both imaging and sequencing for the same single cells. We recently developed an automated system of single cell picking and sequencing. This makes it possible to observe and pick single cells under a microscope and to sequence them in a high throughput manner, which may link the dynamic behavior and whole gene expression for many cells. We will discuss its applications for Singularity Biology.

2SCP-5 (2Pos147) Morphodynamic feature space of migrating cells

Daisuke Imoto[1], Nen Saito[2], **Satoshi Sawai**[1,3] ([1]*Graduate School of Arts and Sciences, University of Tokyo*, [2]*Universal Biology Institute, Graduate School of Science, University of Tokyo*, [3]*Research Center for Complex Systems Biology, University of Tokyo*)

We will introduce a minimalistic model that describes the essential regulatory logics underlying complex morphology of amoeboid cells. The proposed model recapitulates the overall cell forms observed in freely migrating Dictyostelium, neutrophils and fish keratocyte. Similarities between the simulated data and real cell data were assessed by feature extraction by deep learning. Our numerical analyses suggest importance of speed of membrane protrusion and the total amount of key positive regulators of membrane protrusion. Theoretical analysis will be presented along with demonstration of applicability of our approach to analyzing the migratory response in chemotattractant field and cell-cell adhesion.

2SCP-6 (2Pos243) 上皮メカノケミカル動態の同定
(2Pos243) System identification of mechano-chemical epithelial sheet dynamics

Yoshifumi Asakura[1], Yohei Kondo[2], Kazuhiro Aoki[2], Naoki Honda[1] ([1]*Grad. Sch. Biostudies, Univ. Kyoto*, [2]*Div. Quantitative Biol. ExCELLS, NIBB.*)

Collective migration of epithelial cells is a fundamental process of multi-cellular organisms. Our recent study using live imaging with FRET-based biosensor discovered that cell migration within an epithelial sheet is oriented by traveling waves of ERK activation. However, how the cells make a decision on migration direction by integrating mechano-chemical signals has remained still elusive. Here, we performed reverse-engineering approach to extract a hidden control role in the epithelial sheet dynamics in a data-driven manner. Our model has an ability to forecast cell migration quantified in time-lapse images. Therefore, our approach would be powerful to understand mechano-chemical epithelial sheet dynamics.

2SDP-1 クライオ電子顕微鏡による繊毛と微小管モーターの解析
Cryo-EM analysis of cilia and microtubule-based motor proteins

Masahide Kikkawa (*The Univ. of Tokyo*)

Eukaryotic cilia are complex cell organelles and play important roles in various cell, such as propeller and antenna. They consist of hundreds of different proteins that are precisely organized by self-assembly mechanisms and are constructed and driven by microtubule-based motors. To explore the complex cilia system, we combine genetics and cryo-electron microscopy. For example, we knocked-out efcab1 in zebrafish, which encode calaxin, and found that the mutant zebrafish have situs inversus due to the irregular ciliary beating of Kuppfer's vesicle cilia. We also analyzed the complexes of dynein microtubule binding domain as well as kinesin together with microtubule using high-resolution cryo-EM and elucidated the important bind-unbound switching mechanisms of the motors.

2SDP-2 Cryo-EM Analysis of a Feline Coronavirus Spike Protein Reveals a Unique Structure and Camouflaging Glycans

Tzu-Jing Yang[1,2], Yen-Chen Chang[1,3], Tzu-Ping Ko[1], Piotr Draczkowski[1], Yu-Chun Chien[1,2], Yuan-Chih Chang[4], Kuen-Phon Wu[1], Kay-Hooi Khoo[1,2], Hui-Wen Chang[3], **Shang-Te Danny Hsu**[1,2] ([1]*Institute of Biological Chemistry, Academia Sinica*, [2]*Institute of Biochemical Sciences, National Taiwan University*, [3]*School of Veterinary Medicine, National Taiwan University*, [4]*Institute of Cellular and Organismic Biology, Academia Sinica*)

Feline infectious peritonitis virus (FIPV) is an alphacoronavirus that causes nearly 100% mortality rate without effective treatments. Here we report a 3.3 Å cryoEM structure of an FIPV spike protein, responsible for host recognition and viral entry. Mass spectrometry provided site-specific compositions of high-mannose and complex type N-glycans that account for 1/4 of the total molecular mass, most of which were directly visualized by cryoEM. The N-glycans that wedge between two galectin-like domains within the S1 subunit result in a unique propeller-like conformation for the FIPV spike protein compared to all known homologs of other coronaviruses. The structural insights provide a blueprint for a better molecular understanding of the pathogenesis of FIP.

2SDP-3 CryoEM studies of bacterial glutamine synthetase

Kuen-Phon Wu, Chia-Wei Chou (*Institute of Biological Chemistry, Academia Sinica*)

Bacterial glutamine synthetase (GS) is a dodecameric enzyme with total MW 625 kDa. It responds in cellular glutamine biosynthesis as well as nitrogen fixation. Catalytic reactions of bacterial GS had been revealed in paste decades by combining experimental data from 4 GS species, however, discrepancies exist. Moreover, inhibitor development of bacterial GS is one powerful strategy to stop infections (e.g Tuberculosis) as its bifunctional roles will be blocked simultaneously. At here we determine structural transitions in the catalytic cascade by cryoEM. We have determined 5 cryoEM maps of E. coli GS maps at 2.6-3.3 Å representing distinct reaction states. The structures unveil mechanisms of recruiting magnesium, consuming ATP and converting glutamine from glutamate.

2SDP-4 V 型 ATP 合成酵素の膜内在性ドメイン V_o の単粒子解析
Single particle analysis of membrane embedded domain V_o of V-type ATP synthase

Jun-ichi Kishikawa[1], Takayuki Kato[2], Aya Furuta[1], Atsuko Nakanishi[1], Kaoru Mitsuoka[3], Ken Yokoyama[1] ([1]*Dept. Mol. Biosci., Kyoto Sangyo Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Res. Ctr. UHVEM., Osaka Univ.*)

V-type ATP synthase is composed of hydrophilic V_1 and membrane-embedded V_o. Proton translocation in V_o and ATP synthesis in V_1 is coupled by rotation of the central rotor. Previously, we reported that three density maps of the holo-enzyme, distinguished by the difference in direction of the central rotor, by single particle analysis. In this study, we focused on structural analysis of V_o to improve its resolution. EM images of V_o incorporated into nanodisc were collected by CRYOARM equipped with K2 summit detector and processed by RELION. The obtained V_o map at near atomic resolution clearly indicates that the hydrophilic stator domain was in contact with the rotor subunit. This suggests that the V_o adopts an autoinhibited form, when V_1 dissociates from V_o.

2SDP-5 胃プロトンポンプの輸送機構に対する構造基盤
Structural basis for the transport mechanism of the gastric proton pump

Kazuhiro Abe[1,2] ([1]*Cellular and Structural Physiology Institute, Nagoya Univ.*, [2]*Grad. Sch. Pharm. Sci., Nagoya Univ.*)

The gastric proton pump, H+,K+-ATPase is responsible for acidifying the gastric juice up to pH 1, and is thus an important drug target for treating gastric acid-related diseases. Here we show the crystal structures of the H+,K+-ATPase in tow different conformations. The luminal-open E2P structure revealed a conserved lysine residue that points to the juxtaposed carboxyl residues in the cation-binding site. The unusual configuration of the cation-binding site enables the extrusion of a single proton into the pH1 solution of the stomach. We found a single K+ occluded in the E2-P transition state. This fulfills the energy requirement for the generation of a million-fold proton gradient across the membrane, the highest known cation gradient in any mammalian tissue.

2SDP-6 PAD4 regulates p53 function through protein citrullination

Chien-Yun Lee[1], Guang-Yaw Liu[2], **Hui-Chih Hung**[1] ([1]*National Chung-Hsing University*, [2]*Chung Shan Medical University*)

Peptidylarginine deiminase 4 (PAD4)-catalyzed citrullination has been demonstrated its emerging role as a potential cancer therapeutic target. Deregulation of PAD4 has been found in many types of cancer, implying citrullination levels are highly associated with tumorigenesis. In this study, we reported p53 protein can be citrullinated by PAD4 in vitro and in the cell. The structural and functional analysis demonstrated that citrullination has detrimental effects on p53 tetramerization and thus impairs their DNA-binding ability. In conclusion, this study reported a direct evidence of the PAD4-p53 interaction and a potential mechanism of how PAD4 involves in p53 target gene regulation by citrullinating p53, which strengthen the link between PAD4 and tumorigenesis.

2SEP-1 Materials chemistry of photo-excited triplet state for dynamic nuclear polarization

Nobuhiro Yanai[1,2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*JST-PRESTO*)

Nuclear magnetic resonance (NMR) spectroscopy and magnetic resonance imaging (MRI) are powerful and versatile methods in modern chemistry and biology fields. Nevertheless, they suffer from intrinsically limited sensitivity due to the low nuclear spin polarization at ambient temperature. Dynamic nuclear polarization based on photo-excited triplet (triplet-DNP) is promising since it allows the hyperpolarization at room temperature. Here we introduce our recent progress to make triplet-DNP more accessible and feasible. We show the first example of triplet-DNP of nanoporous metal-organic frameworks (MOFs) to allow the accommodation of biology-relevant target molecules. We also show the first example of air-stable and high-performance polarizing agent for triplet-DNP.

2SEP-2 溶媒和の量子化学効果の分子動力学シミュレーションの開発と生体分子への応用
Incorporation of qunatum chemical effect of solvation into molecular dynamics simulation and the applications to biomolecules

Hiroshi Watanabe[1,2] ([1]*Keio Univ. KQCC*, [2]*PRESTO JST*)

Water molecules entering and leaving proteins are closely related to the protein functions. Quantum chemical effects play important poles in dynamics of such internal water molecules. However, the present molecular simulations cannot incorporate these effects into dynamics analysis because of solvent diffusion. To overcome the problems, we proposed a new computational technique, size-consistent multi-partitioning (SCMP) method, and successfully demonstrated that the SCMP method can achieve stable simulations by effectively taking into account quantum chemical effects of solvation. In the presentation, I would like to introduce the basic concept and the recent progress in further development of the SCMP method, and also present some successful applications.

2SEP-3 多光子顕微鏡の応用による脳組織内の分子動態の可視化解析
Imaging dynamics of molecules inside the brain tissue by the application of multiphoton microscopy

Mutsuo Nuriya[1,2,3] ([1]*Keio University School of Medicine*, [2]*JST PRESTO*, [3]*Yokohama National University*)

Dynamics of molecules inside the brain are the key determinants of its physiology and pathophysiology. Thus, in order to gain further insights, visualization of molecules inside the brain tissue is critical. Multiphoton microscopy realizes deep tissue imaging with high three-dimensional resolution, thus is ideal for such imaging analyses. In addition to the well-known two-photon excitation of fluorescence molecules, other multiphoton phenomena exist and can be applied to various biophysical researches. In this presentation, I will introduce our attempts to utilize multimodal multiphoton microscopy to reveal dynamics of molecules inside the living brain tissues.

2SEP-4 Contextuality and Non-Locality in Quantum Physics and Cognitive Science

Yoshihiro Maruyama (*Kyoto University*)

No-Go theorems in quantum theory, such as Bell's and Kochen-Specker's, tell us that fundamental reality is inherently contextual and nonlocal. Bell-type inequalities may be reformulated so as to be applicable in cognitive science, and they are actually violated in certain cognitive experiments. Does it show human reason is contextual as well as fundamental reality? Do cognitive systems exhibit the same kind of contextuality as quantum systems? Do Bell-type results in cognitive science have such a massive impact on our understanding of the world as those in quantum physics indeed had? In this talk I shall elucidate similarities and dissimilarities between contextuality of reality and contextuality of reason, especially in light of the nature of probabilities involved.

2SEP-5 Label-free molecular vibrational spectro-microscopy

Takuro Ideguchi[1] ([1]*The University of Tokyo*, [2]*PRESTO, JST*)

Fluorescent probes provide molecular selectivity in microscopy at the expense of some drawbacks such as photo-bleaching or cytotoxicity. Vibrational spectro-microscopy is label-free counterpart in molecular-sensitive techniques, where we can have image contrast on molecular vibrations without staining specimens. I will introduce advanced vibrational spectro-microscopy techniques based on Raman scattering or infrared absorption which provide high-speed, high-spatial resolution, or high-sensitivity.

2SEP-6 Nanoscale thermometry and magnetometry in biology using NV center in diamond

Hitoshi Ishiwata[1,2] ([1]*PRESTO*, [2]*Tokyo Institute of Technology*)

Nitrogen-Vacancy (NV) center in diamond allows nanoscale magnetometry and thermometry for analysis of biological process at subcellular level. High sensitivity of NV center enables novel magnetometry technique such as Nanoscale NMR for localized NMR analysis from ~(5nm)3 volume. Quantum thermometry has also been demonstrated with sub-degree temperature resolution by detection of the zero-field splitting for the NV center. Detection of the nuclear spin combined with a thermometer capable of sub-degree temperature resolution provide a powerful new tool in many areas of biological research. In this talk, I will focus on describing biological process that could only be observed using NV center and its extended applications.

2SEP-7 (2Pos288) グラフェン電界効果トランジスタとフェムトリットルチャンバーを用いたデバイ遮蔽を超える電気的バイオセンシング
(2Pos288) Electrical Biosensing beyond the Debye Screening Length Using Graphene Field-Effect Transistor in Femtoliter Microchamber

Takao Ono[1], Yasushi Kanai[1], Koichi Inoue[1], Yohei Watanabe[2], Shin-ichi Nakakita[3], Toshio Kawahara[4], Yasuo Suzuki[4], Kazuhiko Matsumoto[1] ([1]*ISIR, Osaka Univ.*, [2]*Kyoto Pref. Univ of Med.*, [3]*Kagawa Univ.*, [4]*Chubu Univ.*)

Graphene is 2D material which has the highest carrier mobility in known materials. Graphene field-effect transistor (G-FET) has a potential for electrical biosensing, because it transduces the carrier modulation by charged targets to high conductivity change. However, in physiological ionic strength, electrical field around the target is attenuated within 1 nm, due to Debye screening. Here we show an electrical biosensing utilizing enzymatic products of the target, which diffuse and reach the graphene surface. 1) Encapsulated into microdroplet to accumulate the products, G-FET monitored ammonia production of urease in real time. *Helocobacter pylori* was detected owing to its urease reaction in sensitivity below single bacterium. 1) T. Ono *et al., Nano Lett., accepted*.

2SFP-1 共生進化生物学の最前線
Frontiers in experimental evolutionary biology of symbiosis

Takema Fukatsu (*AIST*)

Many insects and other organisms constantly harbor microorganisms inside their body. Due to the close spatial proximity, extremely intimate biological interactions and interdependency are commonly found between the partners called host and symbiont. Novel biological properties are often generated through such associations. In many cases, host and symbiont are integrated into an almost inseparable biological entity. Our main research targets are diverse microbial symbiotic interactions found in insects. Here I present an overview of our research topics on the symbiosis and evolution in insects, particularly highlighting experimental evolutionary approaches to insect-bacterium symbiotic associations.

2SFP-2 What makes animal embryos to follow the hourglass model?

Naoki Irie[1,2], Yui Uchida[1,2], Masahiro Uesaka[3] ([1]*Univ. Tokyo, Sch. of Science*, [2]*Univ. Tokyo, Universal Biology Institute*, [3]*RIKEN*)

Considering the cascade-like flow and accumulation of information during development, upstream or earlier developmental processes seems to be more important, and thus sounds reasonable to be evolutionarily conserved. However, recent studies clarified that mid-embryonic stages, rather than earliest stages are conserved the most during evolution. We tackled how this general tendency appear, and found that negative selection by embryonic lethality does not explain well, but intrinsic features such as fluctuation, and/or pleiotropic constraint may have contributed behind the hourglass-like conservation.

2SFP-3 Impact of polyploidy on the evolutionary rate

Ryudo Ohbayashi[1], Tetsuhiro Hatakeyama[2] ([1]*BDR., RIKEN*, [2]*Dept. of Basic Sci., Univ. of Tokyo*)

Diversification of the number of homologous chromosomes (ploidy) is often observed in a wide range of organisms, i.e., bacteria, archaea, and eukaryotes. In bacteria, although most of the model organisms such as E. coli possess only single chromosome per cell, some species maintain multiple copies of that (polyploid). In spite of its universality, little is known about the importance of polyploidy in evolution. To clarify the impact of polyploidy on the evolution, we first investigated the regulation mechanism of replication of multiple genomes. Then, we revealed the associated inheritance pattern of polyploid and constructed the mathematical model for evolution based on observations. In this symposium, we also discuss our ongoing experiments on artificial evolution.

2SFP-4 Analysis of Evolutionary Constraints and Plasticity by Microbial Laboratory Evolution

Chikara Furusawa[1,2] ([1]*BDR, RIKEN*, [2]*UBI, Univ. Tokyo*)

To understand how the course of evolution is constrained in high-dimensional phenotype and genotype spaces, we performed laboratory evolution under various different stress environments, and changes in phenotypes and genome sequence were analyzed. The results demonstrated that the expression changes were restricted to low-dimensional dynamics, while diverse genomic changes can contribute to similar phenotypic changes. Based on these results, we will discuss the nature of phenotypic plasticity and constraint in bacterial evolution, and possible strategies to predict and control the evolutionary dynamics.

2SGP-1 拡張アンサンブル法を用いたタンパク質-リガンド結合ポーズの自由エネルギー解析
Free-energy analysis of protein-ligand binding pose using generalized ensemble methods

Hiraku Oshima, Suyong Re, Yuji Sugita (*RIKEN BDR*)

Accurate prediction of protein-ligand binding poses and affinities has been a central challenge in computational chemistry. The free-energy calculation based on molecular dynamics simulations is the most rigorous method for predicting the binding poses. However, the simulations are often trapped in local minima of the free-energy surface, causing the slow convergence. Here, we propose a pose prediction method based on the generalized replica exchange with solute tempering. The protein-ligand interaction is weakened at high temperatures, accelerating the sampling of multiple poses. Using the free energy perturbation method, we calculated binding affinities of obtained poses, showing excellent agreement between the calculated and experimental results.

2SGP-2 An efficient screening, an accurate evaluation, and a simple prediction of protein complex structures

Kazuhiro Takemura, Akio Kitao (*Sch. Life Sci. Tech., Tokyo Tech.*)

Through "Priority issue 1 on Post-K computer", we have developed various methods toward accurate and efficient predictions for protein complex structures. Our simple clustering approach provides an efficient screening of docking models by rigid-body dockings. The selected complex models can be accurately evaluated by evaluation with the Energy Representation method of docking generated decoys (evERdock), which calculates the binding free energy difference using solution theory in the energy representation. We also developed Concentrated ligand Docking (ColDock) for protein-ligand complex structure predictions using MD simulation at high ligand concentration.

2SGP-3 (2Pos029) Determination of protonated states for native and mutant structures of HIV-1 protease with indinavir by free energy calculations

Masahiko Taguchi, Ryo Oyama, Masahiro Kaneso, Shigehiko Hayashi (*Kyoto University*)

HIV-1 protease is a target of drug development and various inhibitors binding to two aspartic acids in the catalytic site were developed. However, it has a serious drug-resistance. To resolve the drug-resistance problem, it is importnat to determine protonated states of the two aspartic acids. For one of typical inhibitors, indinavir, it is known that mutation (V82T/I84V) greatly affects its binding free energy. To clarify the structural difference between native and mutant proteases with indinavir, we performed QM/MM free energy calculations. Then we succeeded to determine protonated states for both structures precisely, and revealed a change of hydrated environment around catalytic site by the mutation.

2SGP-4 創薬標的タンパク質の溶液構造解析
Ligand-bound forms of drug-discovery target protein in solution studied by molecular dynamics simulations

Toru Ekimoto[1], Takafumi Kudo[1], Tsutomu Yamane[1], Mitsunori Ikeguchi[1,2] ([1]*Yokohama City Univ.*, [2]*Riken*)

Vitamin D receptor ligand-binding domain (VDR) undergoes conformational change upon a ligand binding. Because the onformational change of VDR depends on kinds of activity of ligands, understandings of the ligand-bound conformations itself and differences of conformations between agonist and antagonist-bound state are important for constructing structure-activity relationship model. Using molecular dynamics simulations, we elucidate solution structures of an antagonist-bound VDR by a combination analysis of Markov state model and small-angle x-ray scattering, and execute structure samplings of agonist- and antagonist-bound VDRs by the generalized REST implemented in GENESIS (Kamiya et al., JCP 2018).

2SGP-5 アラニン置換による抗体親和性の向上のメカニズム
Mechanism of antibody-affinity enhancement through alanine-substitution

Takefumi Yamashita (*The University of Tokyo*)

Antibodies that specifically and strongly bind to proteins expressed on the cancer cell can be used for cancer treatments. Thus, the antibody affinity enhancement is an important issue in the antibody drug development. In this study, we investigate affinity-enhancement mechanisms of a cancer-targeted antibody, B5209B, by using all-atom molecular dynamics (MD) simulations. Recently, it was found that two alanine substitutions enhanced the affinity of B5209B in different by different mechanisms. One enhances the enthalpy contribution and the other enhances the entropy contribution. Our MD simulations clearly shows the atomic-level mechanisms by which alanine enhances the antibody affinity.

2SGP-6 タンパク質アポ構造から出発した発展的分子動力学シミュレーションによる薬剤結合モードの予測
Protein-drug binding mode prediction from the apo-protein structure using a molecular dynamics-based pocket generation approach

Mitsugu Araki[1,2], Yasushi Okuno[1,2] ([1]*Grad. Sch. of Med., Kyoto Univ.*, [2]*RIKEN, AICS*)

Protein-drug binding mode prediction from the apo-protein structure is challenging because drug binding often induces significant protein conformational changes. In this study, we develop a computational workflow that incorporates a novel pocket generation method, and this workflow is applied to CDK2 kinase, which has an especially-closed ATP-binding pocket in the apo-form, and its inhibitors. Compared to a conventional prediction protocol, the prediction accuracy is significantly improved by preceding pocket expansion, leading to generation of conformationally-diverse binding mode candidates.

2SGP-7 Reinforcement Learning and Global Optimization Techniques in Molecular Dynamics Simulations

Kei Terayama[1,2,3], Yasushi Okuno[3], Koji Tsuda[1,4,5] ([1]*AIP, RIKEN*, [2]*MIH, RIKEN*, [3]*Grad. Sch. Med., Kyoto Univ.*, [4]*Grad. Sch. Frontier Sci., Univ. Tokyo*, [5]*NIMS*)

In recent years, we have developed novel approaches for drug-like molecule design, MD-based effective conformation sampling, and acceleration of MD from the viewpoint of information science. At the center of these approaches are the reinforcement learning and various global optimization algorithms. In this talk, we would like to introduce their basic idea and how they can be applied in the field of biophysics including MD simulations.

2SGP-8 (2Pos075) 天然変性タンパク質 p53 を標的としたペプチドの人工設計ー液液相分離の制御ー
(2Pos075) Rational design of peptide targeting intrinsically disordered protein p53 -regulation of function and phase-phase separation-

Kiyoto Kamagata[1], Eriko Mano[1], Yuji Itoh[1], Saori Kanbayashi[1], Masaya Honda[1], Ryo Kitahara[2], Tomoshi Kameda[3] ([1]*IMRAM, Tohoku Univ.*, [2]*Coll. Pharmacy Sci., Ritsumeikan Univ.*, [3]*AIRC, AIST*)

Intrinsically disordered regions (IDRs) of proteins are involved in many diseases. However, the flexible IDRs hinders the use of 3D structure-based design methods. Here, we developed a rational design method to obtain a peptide that can bind an IDR using only sequence information. We applied the method to the disordered C-terminal domain of the tumor suppressor p53. The designed peptide showed the suppression of binding to DNA and sliding along DNA by p53 based on NMR, MD, and single molecule experiments. Also, we found that this peptide suppressed liquid liquid phase separation of p53 by blocking the interaction of the IDRs. Sequence-based design may be useful in designing peptides that target IDRs for therapeutic purposes.

2SHP-1 Deciphering genome organization and dynamics by mathematical modeling and simulation

Soya Shinkai (*RIKEN BDR*)

Genomic DNA is packed into the cell nucleus and highly organized in 3-dimension (3D). Recent genome-wide and high-throughput chromosome conformation capture (Hi-C) technologies have uncovered the 3D genome organization in the last decade. The proper expression of genes requires the orchestration of their regulatory elements within the 3D genome structures. Furthermore, as observed in live-cell imaging experiments, genomes are functional in 4D. To understand these genome data, mathematical modeling in terms of polymer physics is a powerful framework. I will talk about our recent studies on deciphering genome organization and dynamics by mathematical modeling and simulation, which allows for extracting physical insights behind the data.

2SHP-2 Reading out G-quadruplex RNA structure using transient state (TRAST) of photochemical reaction of fluorophores

Akira Kitamura[1,2] ([1]*Lab. Mol. Cell Dynamics, Fac. Adv. Life Sci., Hokkaido Univ.*, [2]*JSPS Scientist for Joint International Research*)

Guanine (G)-rich sequences in nucleic acids such as GGGGCC repeats are prone to assemble into four-stranded structures, called G-quadruplexes. However, due to methodological constraints, evidence of specific structures of such GGGGCC repeats in vivo has been reported not so well. In order to readout G-quadruplexes structures of fluorophore-labeled RNAs, we here introduce a strategy using transient states (TRAST) monitoring of fluorophores depending on local viscosity, sterical constraints, and approximation of guanine residues. TRAST monitoring can be available to readout the RNA structures using changes of such states not only in solution but also in living cells.

2SHP-3 Isolation and analysis of specific cells, organelles and supramolecular complexes using microfluidic microdroplets

Ryo Iizuka (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

Microfluidics allows ultrarapid production of monodisperse microdroplets such as water-in-oil microdroplets. The resultant microdroplets are referred to as microfluidic microdroplets. Each microdroplet serves as a nano/picoliter-volume test tube, which is suitable for handling of single cells and single molecules. Thus, microfluidic microdroplets have been used in various applications including single cell genomics and transcriptomics, digital PCR, and directed enzyme evolution. In this symposium, I will introduce our trials for the isolation and analysis of specific cells, organelles, and intracellular supramolecular complexes using microfluidic microdroplets. These techniques will be powerful and efficient tools for decoding intracellular architecture.

2SHP-4 (2Pos268) 大気圧走査電子顕微鏡 ASEM による骨組織再構築の水中免疫電顕法と cryo-TEM 観察
(2Pos268) Observation of unstained bone tissues and immuno-EM in liquid by ASEM and cryo-TEM

Chikara Sato[1], Shinya Sugimoto[2], Yuri Hatano[1], Mari Sato[1], Eiko Sakai[3] ([1]*Biomedical Res. Inst., AIST*, [2]*Dept. Bacteriol., The Jikei Univ. Sch. Med.*, [3]*Dental Pharmacology, Nagasaki Univ.*)

ASEM observed aldehyde-fixed bone tissue mineralization in natural aqueous buffer, decreasing the risk of changes. Mineralization trafficking in osteoblasts and trabeculae of mineralized femur spongy bone surrounded by cortical bone were directly visible. Associated large and small cells revealed by PTA staining suggest remodeling by bone-forming osteoblasts and bone-absorbing osteoclasts, confirmed by immuno-EM in solution targeting cathepsin-K protease digesting collagen fibers in bone[1]. Axonal segmentation controlling neuron trafficking was visualized[2]. MRSA biofilm formation was monitored by ASEM[3] and cryo-TEM. [1]Sato et al. Scientific Reports 9, 7352, 1-13 (2019). [2]Kinoshita et al. Sci Rep 7,41455,1-14 (2017). [3]Sugimoto et al. Sci Rep 6,25889,1-13 (2016)

2SHP-5 (2Pos246) 細胞内のインスリン様成長因子-I(IGF-I)シグナルは振動する
(2Pos246) Cellular insulin-like growth factor-I (IGF-I) signal can be oscillated

Masato Masuda, Fumihiko Hakuno, Shin-Ichiro Takahashi (*Dep. App. Ani. Sci., Grad. Sch. Agr. Lif. Sci., The Univ. Tokyo*)

Insulin-like growth factor (IGF)-I, whose structure is similar to proinsulin, is well known to be secreted constantly, and to mediate long-term actions such as cell growth, differentiation and survival. Recently we have reported studies on the detailed regulatory mechanism of IGF-I signaling including feedback loop. Based on these results, we made a numerical model to simulate the IGF-I signaling in the constant presence of IGF-I. Simulation results and Routh-Hurwitz stability criterion of this model indicated that the IGF-I signal is shown to be stable when IGF-I concentration is high, whereas the IGF-I signal is oscillated when IGF-I concentration is low. Our analysis demonstrated that IGF-I signal can be oscillated in each cell in the constant presence of IGF-I.

2SHP-6 入力制御によるライブセルイメージング解析
Live cell imaging analyses by input control system

Kazuya Kabayama[1,2,3] ([1]*Department of Chemistry, Graduate School of Science, Osaka University*, [2]*Project Research Center, Graduate School of Science, Osaka University*, [3]*Institute for Radiation Sciences, Osaka Univeresity*)

In recent years, live imaging of immune cells has been desired. However, in the case of floating cells, behavior analysis is not easy due to focal shift or movement out of the observation area. Therefore, we established a nano-wrapping technology to keep suspended cells in the observation area by using an ultrathin polymer film with thickness in the tens of nanometers made of polylactic acid. And, we also focused on imaging at physiological concentrations of the innate immune ligand and succeeded in observing receptor-dependent internalization. In addition, we report cell membrane fluidity measurements by introducing volatile anesthetics using a microfluidic device.

3SBA-1 Theoretical model of the allosteric transition of oxygen sensor domain of FixL

Takahisa Yamato (*Nagoya University*)

The effects of ligand binding on an oxygen sensor protein, FixLH, were investigated with the energy exchange network (EEN) model in which residue interactions were evaluated in terms of local transport coefficients of energy flow. As a result, the difference map of EEN between the deoxy and oxy (deoxy and carbomonooxy) states clearly demonstrated the allosteric transition. The difference EEN maps showed that two regions, (A) the junction between the coiled-coil linker and the sensor domain and (B) the potential dimer interface, experienced considerable change of the energy-transport coefficients, indicating that these two regions play important roles in quaternary structural changes and signal transduction in response to ligand binding.

3SBA-2 Molecular Mechanism of NO Reduction by Nitric Oxide Reductase in Cellular System

Yoshitsugu Shiro (*Univ. of Hyogo*)

Nitric oxide reductase (NOR) is a mambrane-integrated and heme-containing enzyme, which is involved in bacterial denitrification under anaerobic system. It catalyzes the reduction reaction of cytotoxic NO (NO-decomposing enzyme), which is produced by NO-generating enzyme, nitrite reducase (NiR), as an intermediate compound of the denitrification. We found that these two enzymes make a complex, which allows a smooth trasnfer of NO from the NiR to NOR. We have also characterized the chemical mechanism of the NO reduction on the active site of NOR, which is a binuclear center consising of the heme and non-heme iron. We can discuss mechanism of the NO decomposition by the denitrification bacteria without diffusion of the cytotoxic NO into the cellular system.

3SBA-3 Direct observation of vibrational energy flow in hemeproteins

Misao Mizuno (*Grad. Sch. Sci., Osaka Univ.*)

Hemeproteins are ideal systems to study vibrational energy flow in proteins. Upon photoexcitation, large excess energy is selectively deposited on heme within 100 fs. The excess energy is redistributed to surrounding protein moiety, then to water. We have conducted picosecond time-resolved anti-Stoked UV resonance Raman spectroscopy on tryptophan residues as a spectroscopic thermometer built in protein moiety. This technique allows us to monitor the excess energy on residue-to-residue basis. Thus, by combination with site-directed mutagenesis, we were able to carry out mapping of energy flow in hemeproteins. Our recent studies suggested that vibrational energy flow in hemeproteins occurs predominantly via van der Waals atomic contacts.

3SBA-4 ミオグロビンとヘモグロビンの結晶内光解離反応過程の観測
Observation of photolysis reaction of myoglobin and hemoglobin in crystals

Ayana Sato-Tomita (*Jichi Med. Univ.*)

Two kinds of techniques are introduced to capture a photolysis intermediate of hemeproteins, CO-bound myoglobin and hemoglobin. The first method is X-ray crystallography combined with laser pumping and cryogenic trapping. The second method is pump-probe X-ray crystallography using pulsed laser and pulsed X-ray of synchrotron radiation. At cryogenic temperatures under 50 K, a photo-dissociated ligand (CO) molecule can move to only the limited range near the heme. By contrast, at 100-140 K under continuous laser irradiation, the ligand migration and protein relaxation can be observed. Around room temperature, we can carry out a real-time measurement to obtain protein reactions.

3SBA-5 Watching energy transport in proteins: Identifying dynamic networks and thermodynamic properties

David Leitner (*University of Nevada, Reno*)

Energy transport in a protein mediates chemical reactions and function. New time-resolved measurements, and a variety of computational methods allow us to map out and describe energy transport in great detail. I will describe some of our computational work on the nature of energy transport in proteins, with focus on what we can learn about protein dynamics and thermodynamics by watching energy flow. By coarse graining energy transport dynamics from the all atom to residue level, we have identified a relation between conformational dynamics at equilibrium and rates of energy transfer across non-bonded contacts. Measurements of rates of energy transfer thus provide a window into equilibrium dynamics of proteins and entropy associated with the dynamics of the contact.

3SCA-1 君子は豹変す: シアノバクテリアも心変わりする
Cyanobacteria change their mind

Daisuke Nakane, Takayuki Nishizaka (*Dept. Phys., Gakushuin Univ.*)

Light is everywhere on the Earth. Even in a small life-form of organism, they sense light direction and controlled their motility to get energy or to avoid damage. Here we demonstrated that rod-shaped unicellular cyanobacteria showed Type-IV-pilus dependent positive phototaxis toward a directional green light, but the cell behavior had changed to negative phototaxis when we applied additional blue light on a short time scale. This reversal of phototactic behavior requires not the directional illumination of blue light, but the total amount of intensity in single cell level. This study provides an insight into how cell optimize information processing with every moment to use light as an energy source in natural environment.

3SCA-2 Visualization of bacteria motility strategies and biofilm formation in tight microfluidic environments

Andrew Utada (*Univ. of Tsukuba*)

Bacteria exist either freely or as members of surface-adhered 3D communities, encased in a slimy extracellular matrix; these aggregates are actually social groups called biofilms. The biofilm lifestyle is fundamental to bacteria biology and ecology, but in many instances the drivers of biofilm formation are unclear. Thus developing tools to aid in the analysis of biofilm formation may enable control and, ultimately, suppression on unwanted surfaces. We describe the development of biofilms in a 2D microfluidic chamber that enables long-term time-lapse microscopy, single-cell tracking, and control over the microenvironment enabling the analysis of the first stages of biofilm formation. We compare and contrast the different strategies of environmental bacteria.

3SCA-3 アーキアべん毛の生物物理学的視点による特性評価
Biophysical characterization of molecular motors in archaea

Yoshiaki Kinosita[1,2], Miller Helen[1], Li Zhengqun[2], Nagisa Mikami[2], E.F. Tessa Quax[2], Sonja-Verena Albers[2], Richard Berry[1] ([1]*Oxford University*, [2]*University of Freiburg*)

Archaea swim by rotating an archaeal flagellum called archaellum. Here, to characterize the motor properties, we applied biophysical techniques and genetics to halophilic archaea. To elucidate the CheY function in archaea, we performed a bead assay in which the motor rotation can be visualized through the bead rotation attached to filament. It demonstrated that an archaeal CheY induced CW rotation like a bacterial CheY. Next, we performed FRAP experiment to determine the kinetics and dynamics of motors. This technique visualized the dynamics of stator proteins, and the recovery half-time was estimated to be approximately 1 min. Now, we are analyzing the effect of ATP turnover on motor exchanges using mutants.

3SCA-4 バクテリア細胞質のガラス的動力学の代謝活動による流動化現象について
Glassy dynamics of a model of bacterial cytoplasm with metabolic activities

Norihiro Oyama[1], Takeshi Kawasaki[2], Hideyuki Mizuno[3], Atsushi Ikeda[3] ([1]*AIST*, [2]*Nagoya University*, [3]*University of Tokyo*)

Recent experiments have revealed that cytoplasms become glassy when their metabolism is suppressed, while they maintain fluidity in a living state. The mechanism of this active fluidization is not clear, especially for bacterial cytoplasms, since they lack traditional motor proteins, which can be described by conventional active matter models, self-propelled particles. We introduce a model of bacterial cytoplasm focusing on the impact of conformational change in proteins due to metabolic activities. In the model, proteins are treated as particles under thermal agitation, and conformation changes are treated as changes in particle volume. Simulations could reproduce experimentally observed nontrivial phenomena, such as active fluidization and fragility change.

3SCA-5 ボルボックス目緑藻の光行動
Photomovements of *Chlamydomonas*, *Volvox* and *Tetrabaena*

Ken-ichi Wakabayashi (*CLS, Tokyo Tech*)

Green algae of the order Volvocales swim with flagella and display photomovements in response to surrounding light. A unicellular species *Chlamydomonas reinhardtii* is the model organism to study regulatory mechanisms of flagellar motion for photomovements. Recently, we found that a large spheroidal species *Volvox rousseletii* shows the same photomovements as *C. reinhardtii* by different flagellar regulation. To understand how the regulatory mechanisms for flagellar motion changed during the evolution of multicellularity, we studied photovements of *Tetrabaena socialis*, the four-celled species. However, *T. socialis* shows only weak photomovements. How *T. socialis* survives in the field will be discussed.

3SCA-6 Collective swimming of living spinners

Azusa Kage[1], Takayuki Torisawa[2,3], Ayano A. Medo[4,5], Ken H. Nagai[6] ([1]*TUT*, [2]*NIG*, [3]*SOKENDAI*, [4]*U Hyogo*, [5]*Present address: Kyoto U*, [6]*JAIST*)

Chlamydomonas reinhardtii is a model organism in flagellar motility. Unlike the wild type swimming with two flagella, the uniflagellated mutant uni1-1 rotates with little translation. We have characterized the collective motion of the living spinners. We found two classes of collective swimming, both of which increased overall translational movement: two-body and many-body swimming. Two-body swimming was realized by the intercellular coupling with flagella, while many-body motion was characterized by hydrodynamic interaction without direct contact between cells. Many-body motion arises from clustering of several spinners. Little translational movement was observed in the spinners at the center of clusters, while the other cells in the cluster showed translation.

3SCA-7 魚類表皮の遊走細胞ケラトサイトのストレスファイバ車輪の回転
Rotation of stress fiber-wheel in migrating fish keratocytes

Chika Okimura (*Fac. Sci., Yamaguchi Univ.*)

Keratocytes are wound-healing motile cells in fish skin. Expansion of the leading edge of keratocytes and retraction of the rear are respectively induced by actin polymerization and contraction of stress fibers in the same way as for other cell types. However, it seems that contraction of stress fibers in keratocytes cannot efficiently retract the rear, because stress fibers in keratocytes align almost perpendicular to the migration-direction. We speculated that the stress fibers may play a role for migration other than the retraction. Here, we reveal that the stress fibers are stereoscopically arranged so as to surround the cytoplasm in the cell body; we directly show, in sequential three-dimensional recordings, their rolling motion as a single wheel during migration.

3SDA-1 オルガネラ・オプトジェネティクス - 細胞内 Ca^{2+} ダイナミクスの光操作
Organelle optogenetics: Optical manipulation of intracellular Ca^{2+} dynamics

Hiromu Yawo[1], Toshifumi Asano[2], Hiroyuki Igarashi[3], Toru Ishizuka[4] ([1]*ISSP, Univ. Tokyo*, [2]*Cell Biol., TMDU*, [3]*Western Univ., Schulich Sch. Med. Dent., Canada*, [4]*Tohoku Univ. Grad. Sch. Life Sci.*)

The Ca^{2+}-dependent signals has been revealed to be a pivotal regulator of various cellular functions and are initiated by two sources: the influx from the extracellular space and the release from the intracellular Ca^{2+} stores, such as endoplasmic/sarcoplasmic reticulum (ER/SR). To manipulate the Ca^{2+} release from ER/SR, we established a new method termed "organelle optogenetics" (Asano et al. *Front Neurosci.* 2018). That is, the Ca^{2+}-permeable channelrhodopsin was specifically targeted to ER/SR in several cell lines. Upon blue/cyan light irradiation Ca^{2+} was immediately released from ER/SR and trigger the subsequent Ca^{2+} mobilization. The organelle optogenetics would reveal the physiological significance of intracellular Ca^{2+} dynamics under spatiotemporal precision.

3SDA-2 光遺伝学ツールとしての光サイクル型動物オプシンの最適化
Engineering of photocyclic animal opsin as a potential optogenetic tool

Takahiro Yamashita (*Grad. Sch. of Sci., Kyoto Univ.*)

Optogenetic tools which can control G protein signaling are useful for the analysis of the intercellular signaling. Because animal opsins regulate G protein activity by light, they are potential optogenetic tools. However, vertebrate visual rhodopsin, most well-studied opsin, has several disadvantages (a lack of abilities to bind all-trans retinal and to deactivate by light). Alternatively, bistable opsins overcome these disadvantages and gain attention. Recently, we discovered a novel opsin which binds all-trans retinal and whose activity is regulated by photocycle. Thus, this opsin can repeatedly control G protein activity. In this talk, I would like to introduce novel photocyclic opsins and discuss their potential as an optogenetic tool.

3SDA-3 (3Pos142) 生体組織への応用が期待される光感度の高いチャネルロドプシン
(3Pos142) Novel optogenetics tool: A light-gated cation channel with high-reactivity to weak light

Shoko Hososhima[1], Shunta Shigemura[1], Hideki Kandori[1], Satoshi Tsunoda[1,2] ([1]*Nagoya Institute of Technology*, [2]*JST, PRESTO*)

Optogenetics has revolutionized the study of experimental biology, making it possible to optically manipulate various biological functions in high temporal and high special resolutions. Especially light-gated ion channels such as ChR2 realized optical excitation when expressed in targeted neurons. Here we report electrophysiological properties and optogenetical application of a light-gated cation channel from cryptophyte, GtCCR4. GtCCR4 exhibits powerful channel activity with a high light sensitivity compared to ChR2. When GtCCR4 was expressed in cultured cortical neurons, successful neuronal firing was observed even by weaker light than required for ChR2 excitation without loss of temporal resolutions, suggesting a potential of GtCCR4 as an ideal neuronal stimulator.

3SDA-4 Optical Switches of Membrane Receptor Activities Using CRY2

Takeaki Ozawa (*Dept. Chem., Univ. Tokyo*)

I will focus on an optogenetic method using an optical dimerizer, cryptochrome2 and its partner protein CIB, to control the trafficking of GPCRs. We switched temporally the interaction between β-arrestin and β2-adrenergic receptor (ADRB2) by external light and found that the duration of theβ-arrestin-ADRB2 interaction determines the trafficking pathway of ADRB2. Remarkably, the phosphorylation of ADRB2 by GRK is unnecessary to trigger clathrin-mediated endocytosis, and β-arrestin interacting with unphosphorylated ADRB2 fails to activate mitogen-activated protein kinase (MAPK) signaling. I will show its further application to controlling different the membrane receptor activities by external light.

3SDA-5 腹側被蓋野の GABA 作動性神経がノンレム睡眠を調節する
VTA-GABA neurons regulate NREM sleep

Akihiro Yamanaka[1,2] ([1]*RIEM, Nagoya University*, [2]*CREST, JST*)

Neural mechanism of sleep/wakefulness regulation is far from understood. Here, we found that glutamic acid decarboxylase 67-positive GABAergic neurons in the ventral tegmental area (VTAGad67+) are a key regulator of non-rapid eye movement (NREM) sleep. VTAGad67+ project to multiple brain areas implicated in sleep/wakefulness regulation. Chemogenetic activation of VTAGad67+ promoted NREM sleep whereas optogenetic inhibition of these induced prompt arousal from NREM sleep but not from REM sleep. VTAGad67+ neurons showed the highest activity in NREM sleep and the lowest activity in REM sleep. VTAGad67+ directly innervated and inhibited wake-promoting orexin neurons. Taken together, we revealed that VTAGad67+ play an important role in the regulation of NREM sleep.

3SDA-6 (1Pos139) 集団細胞遊走における機械的なシグナルを介した ERK 活性伝播
(1Pos139) ERK activation waves mediated by intercellular mechanical signals during collective cell migration

Naoya Hino[1], Xavier Trepat[2], Michiyuki Matsuda[1,3], Tsuyoshi Hirashima[3] ([1]*Grad. Sch. of Biostudies, Kyoto Univ.*, [2]*IBEC, Spain*, [3]*Grad. Sch. of Med., Kyoto Univ.*)

During collective cell migration of epithelial cells, ERK MAP kinase activation propagates as multiple travelling waves from leader cells to follower cells, which orients cell movement. However, how the ERK activation propagates remains elusive. Here we found that intercellular force transmission mediates the ERK activity propagation by using FRET imaging and optogenetic control. Leader cell movement triggers extension of follower cells, resulting in ERK activation in the follower cells. The ERK activation in turn induces cell contraction, which pulls the adjacent cells, evoking another round of extension and ERK activation. Together, we reveal the principle of intercellular communication via mechanical forces underlying long-distance transmission of directional cues.

3SEA-1 単一細胞内温度シグナリングによるストレス顆粒形成の分子機構
The molecular mechanism of thermal signaling-dependent SG formation in single cells

Kohki Okabe[1,2] ([1]*Grad. Sch. Pharma. Sci., The Univ. of Tokyo*, [2]*PRESTO, JST*)

Previously, by measuring and manipulating the temperature in single cells, we showed that the initiation of stress granule (SG) formation depends on intracellular thermal signaling. Here, we investigate the molecular mechanism of this local thermogenesis-dependent cell function. The direct observation of single mRNA particles during stress or locally heating in cells suggested that the local temperature increase in stressed cell destabilize mRNP complex leading to granulation. Next, we revealed the important contribution of microtubules to this thermal signaling of SG formation, which might be due to microtubules-dependent mitochondrial thermogenesis. These results demonstrated that unique interaction of heat and micromodules enables heat-driven signal transduction.

3SEA-2 (1Pos263) ラマンイメージングを用いた細胞内の水の可視化とラベルフリー細胞内温度測定への応用
(1Pos263) Raman imaging of water in a cell and its application to label-free evaluation of intracellular temperature

Toshiki Sugimura, Shinji Kajimoto, Takakazu Nakabayashi (*Grad. Sch. Pham. Sci., Tohoku. Univ*)

We have previously reported quantitative evaluation of water in a cell using Raman microscopy. Water density in nucleus was shown to be higher than that in cytoplasm in a HeLa cell. Here, we applied Raman imaging of water to perform label-free evaluation of intracellular temperature.We measured Raman images of HeLa cells and evaluated the intensity ratio of two regions on the O-H band at different temperatures. The intensity ratio changed linearly with temperature, and the calibration line was obtained both in nucleus and in cytoplasm. We measured the increase in cytoplasm temperature with the addition of FCCP in a cell. A rise in the intracellular temperature was observed, indicating that intracellular can be measured using Raman images of water without staining dye.

3SEA-3 (3Pos064) 温度上昇とテラヘルツ光照射は転写反応に異なる影響を及ぼす。
(3Pos064) Terahertz radiation and temperature increase differently affect transcription by RNA polymerase

Masahiko Imashimizu[1], Masahito Tanaka[1], Hiromichi Hoshina[2], Koh Takeuchi[1] ([1]*AIST*, [2]*RIKEN*)

Thermally driven fluctuations play a critical role in key cellular processes such as DNA replication and transcription. Recent spectroscopic studies have shown that such fluctuating motions of protein and DNA include collective vibrational modes, which are detected as sharp peaks in the terahertz (THz) frequency region. In this study, the effects of THz radiation on transcription by RNA polymerase were analyzed by high-throughput sequencing. We demonstrated that THz radiation enhanced transcription pausing (~kT process), while temperature rise without THz radiation accelerated escape of the paused complex to elongation (>>kT process). This result suggests that the THz radiation specifically affected the biochemical reaction in a manner separable from thermal effects.

3SEA-4 小胞体-ミトコンドリア間クロストークを介した褐色脂肪細胞の機能制御
Regulation of brown adipocyte function through the crosstalk signaling between mitochondria and the endoplasmic reticulum

Hideki Nishitoh (*Lab. of Biochem. and Mol. Biol., Dept. of Med. Sci., Univ. of Miyazaki*)

Mitochondria play a central role in thermogenesis in brown adipocytes (BA), however the molecular mechanism of their quality control system remains unknown. Here we present the importance of mitochondria-ER crosstalk signaling in BA function. The electron microscopic analysis revealed that the ER was surrounded by expanded mitochondria and the area of ER-mitochondria contact site was significantly increased in the differentiated BA. During differentiation, the ER-resident sensor, PERK, is phosphorylated independently of ER stress-induced activation mechanism. Moreover, deletion of PERK impaired the β3 adrenergic receptor-mediated thermogenesis. Our findings suggest that the mitochondria-ER crosstalk signaling-mediated PERK activation is essential for the BA function.

3SEA-5 膜脂質を介する細胞内温度の制御機構
Membrane lipid-mediated regulation of intracellular temperature

Akira Murakami, Kohjiro Nagao, **Masato Umeda** (*Kyoto University*)

Temperature is a fundamental physicochemical parameter that governs chemical reactions and dynamics of functional molecules in cells. Although cell-autonomous control of intracellular temperature is required to counteract the fluctuation of environmental temperatures, it remains unclear how cell recognizes and regulates intracellular temperature. We recently found that low temperature-induced expression of fatty acid desaturase, an enzyme that introduces a double bond into the acyl moiety of lipids, has a crucial role for the regulation of intracellular temperature in Drosophila cells. Molecular mechanisms underlying the membrane lipid-mediated mechanism for regulation of intracellular temperature will be discussed.

3SFA-1 How nano-space affects biological phenomea

Hisashi Tadakuma (*IPR, Osaka University*)

In biological phenomena, related molecules do not associate in dilute solution but react in molecular-crowded condition. Thus, surrounding environment of molecule is important. Moreover, considering the size of biological molecules, the nano scale environment is the key. However, in the past, most of the researches assumed homogeneous environment due to technical limitation. In contrast, recent researches have been revealed that the heterogeneous local environment affects much on the biological reactions. Recently we establish DNA origami-based transcription nano-chip that allows us to evaluate the effect of molecular-layout / environment at nano-meter scale. I will talk our recent results and discuss the intrinsic features of emerging "biophysics in nano-space".

3SFA-2 グアニン四重鎖と i-モチーフ構造を分子プローブとして使ったナノ空間の物性の検討
Investigation of physical properties of a confined nanospace using G-quadruplex and i-motif as a molecular probe

Masayuki Endo (*Grad. Sch. Sci. Kyoto Univ.*)

Here, we examined the influence of the nano-sized space on the physical properties of G-quadruplex (GQ) and i-motif (iM). We created a nano-sized space using DNA origami, and examined mechanical unfolding of the GQ and iM in the nanocages using optical tweezers. We found that the GQ placed in the nanospace was mechanically and thermodynamically stabilized and folding occurred rapidly (Nature Nanotechnol. 2017, 12, 5829). By using four different sizes of nanocages, we found that the stability of GQ and iM increased with decreasing size of nanocages. It was also found that the water activity reduced by decreasing the size of nanocages. This indicates that the stability of iM and GQ in nanocages is correlated with the decrease in water activity (PNAS, 2018, 115, 95392).

3SFA-3 A widespread family of heat-resistant obscure (Hero) proteins protect against protein instability and aggregation

Kotaro Tsuboyama[1], Shintaro Iwasaki[2], Yukihide Tomari[1] ([1]*UTokyo IQB RNA function lab*, [2]*Riken RNA systems biochemical lab*)

We will report that heat-resistant obscure (Hero) proteins, which remain soluble after boiling at 95C, are widespread in Drosophila and humans. Hero proteins are hydrophilic and highly charged, and function to stabilize various “client” proteins, protecting them from denaturation even under stress conditions such as heat shock, desiccation, and exposure to organic solvents. Hero proteins can also block several different types of pathological protein aggregations in vitro, in cells, and in Drosophila strains that model neurodegenerative diseases. Our study reveals that organisms naturally use Hero proteins as molecular shields to stabilize protein functions, highlighting their biotechnological and therapeutic potential.

3SFA-4 有限体積下で働く分子システム設計：人工細胞モデル構築を通して
Molecular system design that works under finite volume: through artificial cell model construction

Shin-ichiro Nomura (*Dep. Robotics, TOHOKU Univ.*)

Biophysical nanospace plays their unique role inside the microspace. The functional reproduction of such nested units of life, so-called, live cell structure, is an interesting problem, and an important research theme as a new understanding of life and a new design guideline for artifacts. Here, as an example of the multimolecular interaction in the lipid membrane-enclosed space, we will introduce the following achievements; the folding effect of DNA Origami, the execution of the DNA molecular amplification circuit, and the result of driving switching of the molecular motor. (these have not been confirmed at present in nanospace!) We would like to discuss the prospects of molecular system design research in micro enclosed space.

3SFA-5 (3Pos195) Intracellular delivery of biologics using magnetically-navigated nanocarrier

Yoshihiro Sasaki, Ryosuke Mizuta, Naoya Kinoshita, Kazunari Akiyoshi (*Department of Polymer Chemistry, Graduate School of Engineering, Kyoto University*)

Various cells secrete exosomes for intercellular communication. They carry RNAs a capable of controlling cellular functions and can be used as drug delivery system nanocarriers. There is the current need to further improve the efficiency of exosome uptake into target cells. Here, we prepared a hybrid of exosomes and magnetic nanoparticles, which could be guided to target cells by a magnetic field for efficient uptake. By applying a magnetic field to the hybrid, exosomes were efficiently transferred intol target cells as conformed by confocal laser microscopy. We also found that differentiation of stem cells to neuron-like cells was enhanced by magnetic induction of the exosome-magnetic nanogel hybrid, indicating maintenance of the intrinsic functions of the exosomes.

3SFA-6 Using SABER to amplify multiplexed FISH signal from RNA and DNA targets

Jocelyn Y Kishi[1,2,5], Sylvain W. Lapan[3,5], Brian J. Beliveau[1,2,5], Emma R. West[3,5], Allen Zhu[1,2], Hiroshi M. Sasaki[1,2], Sinem K. Saka[1,2], Yu Wang[1,2], Constance L. Cepko[3,4], Peng Yin[1,2] ([1]*Wyss Institute, Harvard Univ.*, [2]*Dept. Systems Biology, Harvard Medical School*, [3]*Dept. Genetics, Blavatnik Institute, Harvard Medical School*, [4]*Howard Hughes Medical Institute*, [5]*These authors contributed equally*)

Fluorescent in situ hybridization (FISH) reveals the abundance and positioning of nucleic acid sequences in fixed samples. However, it remains difficult to label more than a few targets and to visualize nucleic acids in environments such as thick tissue using conventional FISH technologies. Here, we introduce signal amplification by exchange reaction (SABER), which endows oligo-based FISH probes with long, single-stranded DNA concatemers that serve as targets for sensitive fluorescent detection. We establish that SABER effectively amplifies the signal of probes targeting nucleic acids in fixed cells and tissues, can be deployed against at least 17 targets simultaneously, and detects mRNAs with high efficiency. SABER is a simple, versatile, and cost-effective method.

3SFA-7 (3Pos075) 三次元構造モデルから発生過程における細胞機能の理解を試みる
(3Pos075) Attempt to understand the cellular function during developmental process from 3D structural model

Junpei Kuroda[1,4], Takeshi Itabashi[1,2,3], Takako M. Ichinose[1], Shigeru Kondo[4], **Atsuko H. Iwane**[1,2,3] ([1]*Cell Field Struc., BDR, Riken*, [2]*Grad. sch. Integ. Sci. Life, Hiroshima Univ.*, [3]*Spec. Res. Promot. Group, Grad. Sch. Fronti., Biosci., Osaka Univ.*, [4]*Pattern formation, Grad. Sch. Fronti., Biosci., Osaka Univ.*)

In order to understand how tissue complex morphology is formed in developmental process, it is necessary to reveal three-dimensionally the positional information and morphology information of individual cells correlated of cell biological analysis. Therefore, we decided to try a method using FIB-SEM and 3D-reconstruction techniques to understand the morphogenesis mechanisms of tissue and the cellular functions in developmental process. We focused on collagen crystals called actinotrichia, which are thought to play a central role in zebrafish fin formation. In this meeting, we will present the interaction between actinotrichia and the cells surrounding it involved in a three-dimensional manner during the fin development, and discuss the cell functions from our 3D models.

3SFA-8 ナノスケール空間における中心小体複製メカニズム
Mechanisms of centriole duplication in nano-space

Daiju Kitagawa, Shohei Yamamoto, Daisuke Takao (*Graduate School of Pharmaceutical Sciences, The University of Tokyo*)

The molecular mechanism that determines a single site for centriole duplication remains a long-standing question. Here, we demonstrate that intrinsic self-organization of Plk4 underlies symmetry breaking in the process of centriole duplication. Our biochemical and cell biological analyses revealed that Plk4 has an ability to phase-separate into condensates via an intrinsically disordered linker and that the condensation properties of Plk4 are regulated by autophosphorylation. We further found that autophosphorylated Plk4 is already distributed as a single focus around the mother centriole before the initiation of procentriole formation. Overall, we propose that the spatial pattern formation of Plk4 is a determinant of a single duplication site per mother centriole.

3SGA-1 Advancing molecular medicine with quantitative single molecule localization microscopy

Devin L. Wakefield[1], Kathleen M. Lennon[1], Steven J. Tobin[1], Matthew S. Brehove[1], Adam L. Maddox[1], Ajay Goel[3], Kendall Van Keuren-Jensen[2], Daniel Schmolze[1], **Tijana Jovanovic-Talisman**[1] ([1]*City of Hope*, [2]*TGen*, [3]*Baylor Research Institute*)

Single molecule localization microscopy (SMLM) can detect single molecules with nanoscale precision. However, it is technically challenging to rigorously quantify receptors in clinical samples: cells from patient tissues and extracellular vesicles secreted by patient cells. To successfully analyze these samples, we tailor our imaging techniques, data analyses, and counting algorithms. We applied our approach to tissue samples from patients with breast cancer; HER2 copy numbers showed a significant positive correlation with detected densities from quantitative SMLM. Additionally, we assessed extracellular vesicles isolated from plasma of patients with pancreatic cancer. We identified a pancreatic cancer-enriched vesicle population and defined vesicle heterogeneity.

3SGA-2 High resolution systems approach to discover mitotic regulation of the nucleus

Paul S. Maddox (*Department of Biology, University of North Carolina at Chapel Hill*)

The nucleus is a complex organelle that ultimately controls cell fate. Each cell division, the nucleus must be disassembled, its genetic material divided equally, and then reassembled. This complex event occurs rapidly and involves numerous protein players. We have probed the biophysical mechanisms that position the nucleus prior to disassembly and thus that govern reassembly. Using high resolution imaging, protein depletions, and computer modeling, we show that regulation of nuclear positioning depends on regulation of microtubule motors and reassembly is achieved by action of balanced protein import and export of visco-elastic regulatory proteins. To further this work, we are developing high-resolution (1.4 NA and higher) light sheet microscopy methodologies.

3SGA-3 Actin-induced compartments and islands in focal adhesions as revealed by simultaneous ultrafast PALM and single-molecule tracking

Takahiro Fujiwara (*WPI-iCeMS, Kyoto Univ.*)

We recently developed an ultrafast camera system, which allowed us to image single fluorescent molecules at a frame rate of 10 kHz with a localization precision of 20 nm, and a 0.33-s resolution live-cell PALM. Virtually all phospholipids and transmembrane proteins were found to undergo non-Brownian hop diffusion between actin-induced ~100-nm-compartments in the plasma membrane (PM), once every ~10 and ~25 ms, respectively. Surprisingly, they also underwent hop diffusion within the focal adhesion (FA) region, where the median compartment size was about half in area. Simultaneous use of ultrafast PALM revealed that integrin molecules diffusing within the FA region intermittently bound to/unbound from FA-protein islands, which might facilitate FA formation-disintegration.

3SGA-4 The axonal cytoskeleton at the nanoscale

Christophe Leterrier (*INP CNRS-AMU UMR7051*)

We use Single Molecule Localization Microscopy (SMLM) to map the nanoscale architecture of actin-based structures within the axon. In the axon initial segment, a key compartment for the maintenance of neuronal polarity, we resolved a highly organized assembly encompassing the periodic actin/spectrin scaffold and its partners: ankyrin, myosin. We have also visualized new actin structures along the axon shaft: rings, hotspots and trails, and are now resolving their molecular organization and functions. For this, we develop a combination of versatile labeling, correlative acquisition and quantitative analysis strategies that allow for high-content, nanoscale interrogation of the axonal architecture.

3SGA-5 (3Pos179) 転写伸長を制御するメディエーターの 1 分子超解像イメージングによる分子局在と動態の定量解析
(3Pos179) Molecular localization and dynamics of Mediator regulating transcription elongation using single-molecule and super-resolution microscopy

Yuma Ito[1], Shinnosuke Kunimi[1], Hidehisa Takahashi[2], **Makio Tokunaga**[1] ([1]*Sch. Life Sci. Tech., Tokyo Inst. Tech.*, [2]*Grad. Sch. Med. Life Sci., Yokohama City Univ.*)

Mediator, a transcriptional coactivator, is a multiprotein complex with multiple functions in transcription activation and chromatin structure alteration. Recently, Mediator is suggested to regulate releasing RNA Polymerase II (Pol II) from promoter-proximal pausing. To elucidate the molecular mechanism, we performed FRAP analysis using domain deletion mutants of Mediator subunit MED26. The results supports a model in which Med26 N-terminal domain plays a role in switching the binding of TFIID to super elongation complex (SEC). We further carried out simultaneous super-resolution and single-molecule imaging of MED26 and Pol II. It enables us to visualize and quantify how molecules interacts with the cluster formed by the other molecules at the molecular level.

3SHA-1 ヨウ素染色によるアミロイド線維構造多形と構造伝播の解析の試み
Iodine staining as a useful probe for amyloid polymorphism and its propagation

Eri Chatani[1], Takato Hiramatsu[1], Keisuke Yuzu[1], Naoki Yamamoto[2] ([1]*Grad. Sch. Sci., Kobe Univ.*, [2]*Fac. Med., Jichi Medical Univ.*)

Amyloid fibrils are associated with amyloidoses and neurodegenerative diseases, and structural details of amyloid fibrils are one of important issues for understanding pathogenesis at molecular levels. Especially, amyloid polymorphism attracts much attention, although its prompt recognition is difficult in many cases because of a lack of sensitive probes. We will talk about our attempts to use iodine staining as a method for distinguishing amyloid polymorphs. By using insulin fibrils, we found that different color tones exhibited depending on polymorphs. Structural propagation by seeding could also be tracked. Based on fundamental properties of the color formation, the availability of the iodine staining as a probe for amyloid polymorphism will be discussed.

3SHA-2 フィブロインタンパク質からクモ糸への人工的再構成
Artificial reconstitution of the multi-hierarchical structure in spider silk from fibroin proteins

Hironari Kamikubo[1,2], Takehiro Sato[3] ([1]*NAIST MS*, [2]*KEK IMSS*, [3]*Spiber Inc.*)

Fibroin protein is a kind of structural proteins, which is a significant component in silk. Notably, the superior properties of spider silk enhance our motivation to utilize it as next-generation industrial materials. The fibroin protein undergoes structural change and regulatory aggregation to form the spider silk, in other words, the “quality” of the fibroin protein dramatically changes in the process of dragline production in the body of a spider. However, the structure formation process is still unclear. In this talk, we would like to introduce the struggle for the artificial reconstruction in vivo of the unique multi-hierarchical structure in spider silk to investigate how the spider fibroin acquires the “quality” of the spider silk.

3SHA-3 Biophysical characterization of environment-dependent protein assemblies of physiological and pathological interest

Maho Yagi-Utsumi, Koichi Kato (*ExCELLS, NINS*)

Proteins are often assembled into filamentous structures, which exert appropriate biological functions, as exemplified by microfilaments, and are deposited as malfunctional aggregates, as illustrated by amyloid fibrils. The filamentous morphology and assembly kinetics critically depend on microenvironments surrounding these proteins. For characterization of their assembling properties, we employ NMR spectroscopy and other advanced biophysical methods with appropriately designed model systems. In this symposium, we will present our biophysical approach to study the molecular mechanisms underlying environment-dependent self-assembling of proteins typified by amyloid-β, using conformational trapping in membrane and microgravity environments.

3SHA-4 (3Pos096) 過渡的に形成される GPCR ダイマーの研究：細胞内蛍光 1 分子観察によるアプローチ
(3Pos096) Examining the transiently formed GPCR dimer: an approach by single fluorescent molecule observation in living cells

Rinshi Kasai (*Inst. Front. Life. Med. Sci., Kyoto Univ.*)

Recent studies have shown that G-protein coupled receptors (GPCRs) are in dynamic equilibrium between monomers and dimers in the plasma membrane. However, the functions and properties of GPCR dimer are not well understood. To investigate them, we performed the single fluorescent molecule observation of GPCR and trimeric G-protein in live cells. As a result, we found that dynamic dimer formation of GPCR as well as transient binding of GPCR dimer to trimeric G-protein were modulated by addition of a ligand or an inverse agonist, an inhibitor of constitutive activity. In particular, it was found that trimeric G-protein recruitment to GPCR dimer depends on constitutive activity, suggesting that transient dimer formation is involved in basal signal production.

3SHA-5 (3Pos014) STAP-2 により Breast tumor kinase が活性化する機構の解明
(3Pos014) Molecular basis of Breast tumor kinase by an adaptor protein, STAP-2

Junki Nakasako[1], Yuki Matsuo[2], Ryo Kanda[2], Yoshino Tanaka[2], Min Yao[3], Tadashi Matsuda[2], Katsumi Maenaka[2], Toyoyuki Ose[2,3,4] ([1]*Graduate school of Life Science*, [2]*Faculty of Pharm.*, [3]*Faculty of Advanced Life Science, Hokkaido University*, [4]*JST PRESTO*)

Breast tumor kinase (Brk) is a nonreceptor tyrosine kinase present in cytosol. Although Brk is expressed frequently in breast cancer cells, usually, it is not observed in normal breast tissue, therefore the possibility of therapeutic application targeting Brk has been pointed out. It has been reported that Brk over-phosphorylates the transcription factors STAT3 and STAT5 through the interacting with signal-transducing adaptor protein-2 (STAP-2) thus causes abnormal cell proliferation. We believe it is important to understand the activation mechanism of Brk. To identify important domains for the activation of Brk, we constructed recombinant expression system of STAT-2 including some mutants. The interaction analysis and enzymatic assay are now now in progress.

3SHA-6 Visualization of Qualitative Change of Proteins with High-Speed Atomic Force Microscopy

Takayuki Uchihashi[1,2] ([1]*Department of Physics*, [2]*ExCELLS, NINS*)

Functions of proteins are elicited through qualitative changes in various physical and chemical properties. One of the important aspects to gain insight of the qualitative change of proteins is dynamics of molecular conformations, assembly states and dynamic interactions. High-speed atomic force microscopy (HS-AFM) has been recently established as a technique enabling visualization of single-molecule dynamics in a physiological condition and thus is a powerful tool to asses quality of proteins from the physical aspects. In this talk, potential of HS-AFM towards studying protein quality will be discussed by demonstrating typical examples associated to the protein quality and function.

3SHA-7 Aggregation and misfolding of therapeutic antibodies in bioprocessing

Masayoshi Onitsuka[1,2] ([1]*Grad. Sch. of Tech. Ind. Soc. Sci., Tokushima Univ.*, [2]*Manufacturing Technology Association of Biologics*)

Mammalian cells are commonly used for industrial manufacturing of therapeutic antibodies as recombinant host cells, and produce and secrete properly folded antibodies with human-like glycosylation. However, the quality of therapeutic antibodies is sometimes problematic. One of the most important issues is the aggregation of antibodies, because of reduced efficacy and immunogenicity. Although studies as to the aggregation of antibodies have been developed in vitro, little is known regarding antibody aggregation in bioprocessing, i.e., multimolecular crowding conditions of intra- and extra-cells during the cell culture process. In this presentation, the mechanism of aggregation and misfolding of antibodies and their structural characterization will be discussed.

1Pos001* HDX-MS を用いた Fc 断片と IgG1 全長の Fc 領域における構造解析
Structural analysis of IgG Fc region in Fc fragment and IgG1 full-body by HDX-MS

Yuki Yamaguchi[1], Tesuo Torisu[1], Susumu Uchiyama[1,2] ([1]*Grad. Sch., Eng., Univ. Osaka*, [2]*ExCELLS*)

Immunoglobulin G (IgG) which is immune related protein is widely used as therapeutic antibodies. IgG is composed of a Fc region and two Fab regions, which are connected through the hinge region. There is possibility that the characters of Fab domain effect on structures and dynamics of Fc domain. However, this hypothesis is still unclear because of the lack of full-length IgG structural information. This study focused on comparison of structures between Fc fragment and IgG1 full-body to examine the effect of the existence of Fab region on structures and dynamics of Fc region in IgG1 using hydrogen/deuterium exchange mass spectrometry (HDX-MS). From HDX-MS results, most of all region in Fc fragment showed higher deuterium exchange rate than IgG1 full-body.

1Pos002* 呼吸鎖における拡張型超複合体の Cryo-EM による構造の解明
Elucidation of the structure of extended super-complex in the respiratory chain by cryo-EM

Kasumi Hirakawa, Wataru Ishibashi, Tomoichirou Kusumoto, Junshi Sakamoto, Takuo Yasunaga (*Grad. Sch Comp. Sci. and Sys. Eng, Kyushu Inst. Tech.*)

Living things produce ATP in the respiratory chain and use it for their activities. In the chain, the electron transfer system is important. Recently, Corynebacterium glutamicum, an amino acid-producing bacterium, has attracted attention not only in the field of food but also in medicine, because it is closely related to pathogenic bacteria. We previously showed the existence of the extended super-complex (complex III/complex IV with NDH-II). Thus we intend to clarify its structure using cryo-EM and single particle analysis, and so we examined purification protocols of monodispersed samples using the adequate detergents. By negatively-staining and cryo-EM, we observed larger particles than complex III/Complex IV. We will here report on the progress.

1Pos003* シゾロドプシンのプロトン輸送の構造基盤
Structural basis of proton transport in Schizorhodopsin

Akimitsu Higuchi[1], Wataru Shihoya[1], Keiichi Inoue[2,3,4,5], Masae Konno[2], Hideki Kandori[2,3], Osamu Nureki[1] ([1]*Department of Biological Sciences, Graduate School of science, University of Tokyo*, [2]*Department of Life Science and Applied Chemistry, Nagoya Institute of Technology*, [3]*OptoBioTechnology Research Center, Nagoya Institute of Technology*, [4]*The Institute for Solid State Physics, The University of Tokyo*, [5]*PRESTO, Japan Science and Technology Agency*)

In 2019, previously-unknown rhodopsin was found within Asgardarchaeota genomes and was named Schizorhodopsin (SzR). SzR phylogenetically locates an intermediate position between Type1 rhodopsins and heliorhodopsins. Because SzR functions as an inward proton pump, SzR might be able to be applied as a new optogenetic tool. For structural analysis, we evaluated multiple SzRs and identified one promising candidate. We purified and crystallized the SzR in lipidic cubic phase (LCP) method. As a result of search for crystalizing condition, we gained the crystal and collected the diffraction data at SPring-8. Here, we report the crystal structure of the SzR at 2.1 Å-resolution. The structure reveals the inward proton pumping mechanism.

1Pos004* カチオン性抗菌ペプチド Hymenochirin-1Pa および変異体 D9K の細菌膜結合構造と膜選択性の解析
Membrane-bound structure and membrane selectivity of cationic antimicrobial peptide Hymenochirin-1Pa and its analog D9K

Akifumi Ohyama[1], Batsaikhan Mijiddorj[2,3], Kazuyoshi Ueda[2], Akira Naito[2], Izuru Kawamura[1,2] ([1]*Grad. Sch. Eng. Sci., Yokohama Natl. Univ*, [2]*Grad. Sch. Eng., Yokohama Natl. Univ*, [3]*Sch. Eng. Appl. Sci., Natl. Univ. Mongolia*)

Cationic antimicrobial peptide Hymenochirin-1Pa (H-1Pa),LKLSPKTKDTLKKVLKGAIKGAIAIASMA-NH2 is isolated from skin secretion of Pseudhymenochirus merlini. H-1Pa shows a strong activity against various bacteria such as methicillin-resistant S. aureus [1]. Our previous research showed Asp9 is possible to be an important role to stabilize membrane-bound structure in the S. aureus membrane. Here, we show membrane-bound structure of H-1Pa and its analog D9K (replacement of Asp9 with Lys9 in the peptide) using by CD, SSNMR and MD simulations. We found the difference of the membrane-bound structure. We will discuss about the effect of Asp9 on membrane-bound structure and membrane selectivity of H-1Pa.
[1] J. M. Conlon et al. (2013) Comp. Biochem. Physiol. D, 8, 352.

1Pos005 (1SCP-5) The role of C-terminal carboxylation in α-conotoxin LsIA interactions with human α7 nicotinic acetylcholine receptor *in silico*

Jierong Wen, Andrew Hung (*Sch. Sci., RMIT Univ.*)

Nicotinic acetylcholine receptors (nAChRs) are ligand-gated ion channels, modulating the signal transmission between neurons. However, their subtype-specific functions are still unclear due to the lack of suitable selective probes. α-Conotoxins are well-known to inhibit nAChRs with high potency, nevertheless, the knowledge of the effects of C-terminus (C-T) modification on subtype selectivity is sparse. In this study, we identified the molecular determinants of α-conotoxin LsIA, with amidated/ carboxylated C-T, binding to human α7 nAChR via molecular dynamics simulations. Results show the atomic-level mechanism of reduced carboxylated LsIA affinity at the α7 versus the amidated LsIA, which may open a new avenue for the design of new leads for nAChR-related diseases.

1Pos006 分子動力学法を用いた Hras-GTP/GDP 複合体の各部の構造変化と各部の水素結合との同時緩和モードの研究
Molecular dynamics study of simultaneous relaxation modes between structures and the hydrogen bonds in the Hras-GTP/GDP complexes

Takeshi Miyakawa[1], Ryota Morikawa[1], Masako Takasu[1,2], Kimikazu Sugimori[2], Kazutomo Kawaguchi[3], Hidemi Nagao[3] ([1]*Sch. of Life Sci., Tokyo Univ. of Pharm. and Life Sci.*, [2]*Inst. of Liberal Arts. & Sci., Kanazawa Univ.*, [3]*Coll. of Sci. and Eng., Kanazawa Univ.*)

In order to understand the role of hydrogen bonds in the Hras-GTP/GDP complexes, 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 simultaneous relaxation mode between the structural deformations of various parts and the hydrogen bonds of various parts in the Hras-GTP/GDP complexes using Ralaxation Mode Analyses.

1Pos007 自動デザイン：分子シミュレーションデータを用いた自動ドラッグデザイン
AutoDesign - an automated drug design by using protein-ligand simulation data

Hironori Kokubo, Naoki Miyamoto, Yoshi Nara (*Axcelead, Inc.*)

Medicinal and computational chemists have struggled for drug design by utilizing various information (Structure-activity relationships, binding models, etc). However, it is very time-consuming to consider many possible design strategies. Recently, performing molecular simulation is getting popular and large-scale data is obtained. But in many cases only partial data is analyzed and utilized for drug design. Here, we propose a novel automated drug design method by using MD simulation data of protein-ligand systems. Our method can automatically interpret complicated simulation data as a rational and comprehensive drug design. We present a basic concept and an application example of drug design by using the data of our original cosolvent simulation (LigMap) and MMGBSA.

1Pos008 Simulating large-amplitude transitions in proteins with a coarse-grained model

Ai Shinobu[1], Chigusa Kobayashi[1], Yasuhiro Matsunaga[2], Yuji Sugita[1,3,4] ([1]*RIKEN Center for Computational Science*, [2]*Saitama Univ., Grad. Sch. Sci. Eng.*, [3]*RIKEN Cluster for Pioneering Research*, [4]*RIKEN Center for Biosystems Dynamics Research*)

Biochemical reactions are often coupled with large-amplitude structural transitions, a very common case is a transitioning from an open to closed state upon substrate binding. Coarse-grained (CG) models are useful for such systems, since they reduce the computational time by several orders of magnitude, allowing access to timescales unreachable by conventional all-atom MD. In this work, we designed a scheme to effectively simulate such transitions using the structure-based CG off-lattice Go model, and implemented it in GENESIS MD software. We applied the scheme to several well-studied systems and show that our mixing model was successful in describing structural transition for proteins undergoing large-amplitude transitions between distant states.

1Pos009 Dynamics and interdomain interactions in a Drosophila adapter protein (Drk) and their correlation to the unfolding of the N-SH3 domain

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Disordered states of proteins are important components of the proteome, wherein they play crucial roles in many cellular processes as well as in human diseases. SH3 domains are marginally stable and exist in equilibrium between folded and unfolded states. Drk and it's human homologue (Grb2) are involved in signal transduction, and consists of two SH3 domains (N-/C- terminal) and a SH2 domain. While N-terminal SH3 fragment exists in 1:1 equilibrium between folded and unfolded states, our recent NMR analysis suggested that such equilibrium is shifted toward total unfolding, in full length Drk. In this study, several micro-seconds molecular dynamics simulations at different conditions were performed to elucidate the interactions of N-SH3 domain in full length Drk and Grb2.

1Pos010 微小管内タンパク質によるチューブリン格子構造の内側からの制御
Microtubule inner proteins regulate the tubulin lattice architecture from the inside

Muneyoshi Ichikawa[1], Ahmad Khalifa[2], Shintaroh Kubo[3], Kaustuv Basu[2], Daniel Dai[2], Amin Maghrebi[2], Javier Vargas[2], Khanh-Huy Bui[2] ([1]*Dept. of Systems Biol., NAIST*, [2]*McGill Univ.*, [3]*Dept. Biophysics, Kyoto Univ.*)

Cilia and flagella oscillate and generate flow of the liquid around eukaryotic cells. Doublet microtubule is a main component of the axoneme structure of cilia and flagella. Despite being composed of same tubulin molecules as singlet microtubules in the cytoplasm, doublet microtubules are highly stable and complex structure with uneven tubulin lattice unlike singlet microtubules. Here, we obtained the first near atomic resolution structure of doublet microtubule by cryo-electron microscopy at 4.3 Å, and complex network of microtubule inner proteins (MIPs) inside the tubulin lattice was revealed. By comparing with microtubule structure lacking some MIPs, the mechanisms of how tubulin lattice architecture is regulated by MIPs from the inside will be discussed.

1Pos011 小角散乱によるアミロイド線維中のヒト α-シヌクレインの構造解析
Structural analysis of human α-synuclein within amyloid fibrils by small-angle scattering

Satoru Fujiwara[1], Tatsuhito Matsuo[1], Yasunobu Sugimoto[2] ([1]*Inst. Quantum Life Science, QST*, [2]*Nagoya Univ.*)

Formation of amyloid fibrils of α-synuclein (αSyn) is closely related to the pathogenesis of Parkinson's disease. Structural analysis of amyloid fibrils is important for elucidation of the mechanism of the fibril formation, and thus elucidation of the mechanism of the pathogenesis. Here we characterize the structure of amyloid fibrils of αSyn by small-angle X-ray and neutron scattering (SAXS and SANS), in particular, the structure of αSyn within fibrils. In addition to the information on the shape and the hydration structure of fibrils obtained from the combined analysis of SAXS and SANS, the information on the structure of individual αSyn within fibrils was obtained using the new SANS method. Comparison with the structure of αSyn in the monomeric state is discussed.

1Pos012 共溶媒の構造類似度を利用した共溶媒分子動力学法における密度マップの類似度の推定
Estimation of the probability map (Pmap) similarity of cosolvent MD (CMD) from structural similarities of cosolvents

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Cosolvent MD (CMD) is an MD simulation of a protein in explicit water molecules mixed with cosolvent molecules. Existing methods utilize small molecules which represent functional groups of compounds, such as isopropanol and benzene. Mahmoud et al. demonstrated that the probability map (Pmap) of a cosolvent atom was highly dependent on its chemical context and suggested that a wider variety of cosolvent molecules should be used in CMD; however, the relationship between the cosolvent structure and the Pmap still remains unclear. Here, we performed CMDs with various cosolvent molecules and analyzed the correlation between the cosolvent structure and the Pmap. Furthermore, we developed a method to predict the change of the Pmap as the change of the cosolvent structure.

1Pos013 単量体タンパク質の長時間シミュレーションのフォールディングパスウェイの動的解析
Dynamical analysis on the folding pathways of long simulations of a single protein

Ayori Mitsutake[1], Hiroshi Takano[2] ([1]*Meiji Univ.*, [2]*Keio Univ.*)

As longer MD simulations are performed, it is more important to develop analysis methods to investigate dynamics and kinetics. We have developed dynamical analysis method, relaxation mode analysis, to investigate “dynamic” properties of structural fluctuations of proteins (for review for biomolecules, see [1]). In RMA, slow relaxation modes are extracted from molecular simulations. This method is powerful to analyze simulations with large structural changes such as folding simulations. We apply RMA to long simulations of a single protein. We discuss the folding pathways of the protein by using RMA.

[1]A. Mitsutake and H. Takano, Biophysical Review, 10, 375-389 (2018).

1Pos014 Targeting the cryptic sites: NMR-based strategy to improve the draggability of proteins by controlling the conformational equilibrium

Koh Takeuchi[1], Yumiko Mizukoshi[2], Yuji Tokunaga[1], Hitomi Matsuo[2], Ichio Shimada[3] ([1]*AIST, molprof*, [2]*JBiC*, [3]*The Univ. Tokyo, Grad Sch Pharm Sci*)

In drug-developments, we experience that ligands bind to pockets that are not evident in the unligated state. These pockets are called cryptic sites and the presence of the cryptic sites usually becomes evident only after the serendipitous finding of the ligands. By using solution NMR, we showed that the cryptic site already exists in the conformational equilibrium of the unligated BclxL, a target of PPI inhibitors. We quantified that ~1% of the unligated BclxL has the cryptic site and an allosteric mutation that stabilizes the cryptic site successfully improved the hit rate of an initial screening. These results indicate that the draggability of proteins can be rationally improved by controlling the population of cryptic site in the conformational equilibrium.

1Pos015 ヘリオロドプシンの構造と生物物理学的解析
Structure and biophysical characterization of the heliorhodopsin

Wataru Shihoya[1], Keiichi Inoue[2,3,4,5], Singh Manish[2], Masae Konno[2], Shoko Hososhima[2], Keitaro Yamashita[1], Kento Ikeda[6], Akimitsu Higuchi[1], Sae Okazaki[1], Izume Tamaki[1], Masanori Hashimoto[2], Ritsu Mizutori[2], Sahoko Tomida[2], Yumeka Yamauchi[2], Rei Abe-Yoshizumi[2], Kota Katayama[2,3], P. Satoshi Tsunoda[2], Mikihiro Shibata[7,8], Yuji Furutani[2,9,10], Alina Pushkarev[11], Oded Beja[11], Takayuki Uchihashi[12,13], Hideki Kandori[2,3], Osamu Nureki[1] ([1]*Department of Biological Sciences, Graduate School of Science, The University of Tokyo*, [2]*Department of Life Science and Applied Chemistry, Nagoya Institute of Technology*, [3]*OptoBioTechnology Research Center, Nagoya Institute of Technology*, [4]*The Institute for Solid State Physics, The University of Tokyo*, [5]*RESTO, Japan Science and Technology Agency*, [6]*School of Mathematical and Physical Sciences, Graduate School of Natural Science & Technology, Kanazawa University*, [7]*Nano Life Science Institute (WPI-NanoLSI), Kanazawa University*, [8]*High-speed AFM for Biological Application Unit, Institute for Frontier Science Initiative, Kanazawa University*, [9]*Department of Life and Coordination-Complex Molecular Science, Institute for Molecular Science, National Institutes of Natural Sciences*, [10]*Department of Structural Molecular Science, The Graduate University for Advanced Studies (SOKENDAI)*, [11]*Israel Institute of Technology*, [12]*Department of Physics, Nagoya University*, [13]*Structural Biology Research Center, Graduate School of Science, Nagoya University*)

Heliorhodopsins (HeR) is a distinct abundant group of microbial rhodopsins recently discovered by metagenomics. Here, we present the crystal structure of the HeR from Thermoplasmatales archaeon. The HeR structure has seven transmembrane helices with an all-trans retinal chromophore, linked to lysine at the seventh transmembrane helix through a protonated Schiff base linkage, as in the type-1 rhodopsins. The linear hydrophobic pocket allows a retinal configuration and isomerization, similar to those in type-1 rhodopsin, whereas most of the residues constituting the pocket are diverged. Our study deepens the understanding of the functions of HeRs, and the structural diversity in the microbial rhodopsins.

1Pos016 クロストリジウム属 2 成分毒素輸送チャネル Ib ポアのクライオ電子顕微鏡構造解析
Cryo-EM structure of clostridial binary toxin translocation channel Ib-pore

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C. perfringens iota toxin is a member of binary toxin, which is composed of membrane binding component (Ib) and actin-specific ADP-ribosyltransferase (Ia). At first, Ib precursor is cleaved off by cellular protease, and then it binds to target cell via receptor, forms soluble oligomer (prepore), and finally forms membrane spanning pore in acidified endosome. Ib-pore acts as a substrate docking platform that subsequently translocates Ia into the cytosol. To understand the translocation channel machinery mechanism, we revealed the structure of Ib-pore at atomic resolution by single particle cryo-EM analysis. We will discuss on the similarities and differences of each channel platform between Ib-pore and anthrax toxin PA-pore in the same group.

1Pos017 藍色細菌 *Anabaena variabilis* 由来 RNA 結合タンパク質の二次構造
Secondary structure of cyanobacterial RNA-binding protein, RbpD, from *Anabaena variabilis*

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RbpD in *Anabena variabilis* is a member of the small RNA-binding proteins (Rbps) containing a single RNA recognition motif. Rbps have the function for the proper control of gene expression, development and stress response, and are regarded as important factors in various organisms. In *A. variabilis*, low temperature induces the expression of *rbp* genes with the exception of *rbpD*. Rbps other than RbpD consist of an N-terminal RRM and a C-terminal glycine-rich domain, and RbpD only contains one RRM domain. To elucidate the functional differences of RRMs between RbpD and other Rbps, we overexpressed the $^{15}N/^{13}C$ labelled RbpD in *E. coli* and solution structure of RbpD was analyzed with multidimensional NMR spectroscopy, and secondary structure of RbpD was determined.

1Pos018 Hydrogen bond donors and acceptors are generally depolarized in α-helices as revealed by a negative fragmentation approach

Yu Takano[1,2], Hiroko X. Kondo[1,3], Ayumi Kusaka[2], Shusuke Yamanaka[4], Nakamura Haruki[2] ([1]*Grad. Sch. Info. Sci., Hiroshima City U.*, [2]*IPR, Osaka U.*, [3]*Faculty Eng. Kitami Inst. Tech.*, [4]*Grad. Sch. Sci., Osaka U.*)

Hydrogen bond (H-bond) is one of the major factors to build protein structures. H-bond energies in α-helices were examined by DFT calculations, followed by a negative fragmentation approach (NFA) to extract a local interaction between molecular fragments. The contribution of each H-bond interaction in α-helices was estimated from NFA, and the results were compared with those in the minimal models, in which only H-bond donor and acceptor exist with the capping methyl groups. The former energies were always weaker than the latter energies. We found that the reduced H-bond energy originated from the depolarizations of both the H-bond donor and acceptor groups, due to the repulsive interactions with the neighboring polar peptide groups (Kondo et al., JCC, in press).

1Pos019 Full atomistic model building of EhV-ATPase using homology modeling/molecular dynamics simulation based on the low resolution cryoEM map

Yu Yamamori[1], Jun Tsunoda[2,3], Ray Burton-Smith[3], Chihong Song[3], Ryouta Iino[2,4], Kazuyoshi Murata[2,3], Kentaro Tomii[1] ([1]*AIST*, [2]*SOKENDAI*, [3]*NIPS*, [4]*IMS*)

The entire structure of EhV-ATPase, an ATP-driven ion pomp in the eubacteria Enterococcus hirae, remains unclear. Recently, single-particle cryo-electron microscopy with Zernike phase plate provided the first structure at 17Å resolution. In this study, we are building an atomic structure model of EhV-ATPase based on the further improved cryo-EM map at subnanometer resolution. Our modeling protocols of EhV-ATPase are as follows. 1) Homology models are built based on the results of profile-profile alignment. 2) Model selection is initially performed by considering the correlations between each model and the cryo-EM map, and finally by structurally evaluating the models. 3) Molecular dynamics simulations are performed with restraint for the density map.

1Pos020 2 次元テンプレートマッチング法によるクロマチン構造多型解析への挑戦
Challenging for multi-conformational analysis of chromatin using the two-dimensional template matching method

Atushi Tokuhisa[1,2,3], Ryo Kanada[1], Shuntaro Chiba[2], Kei Terayama[3,4,5], Shigeyuki Matsumoto[2], Yuta Isaka[1,6], Biao Ma[1,6], Narutoshi Kamiya[7], Yasushi Okuno[1,3,5,6] ([1]*RCH, RIKEN*, [2]*MIH,RIKEN*, [3]*R-CCS,RIKEN*, [4]*AIP,RIKEN*, [5]*Medicine,Kyoto U*, [6]*CCD,FBRI*, [7]*Simulation,U.Hyogo*)

We developed a practical modeling protocol to retrieve a plausible 3D structure of a biomolecule based on a noisy 2D image from single-particle experiment. Our method comprises two procedures, the coarse-grained (CG)-based conformational sampling and an efficient 2D image matching considering many possible molecular orientations. We used nucleosomes, ribosomes and galactosidase as target molecules. At 20-Å resolution, the CG-based template matching method well balanced computational cost and modeling accuracy. Eventually, we succeeded in speeding up the 2D image matching about 100 times by adopting the CG model and Bayesian optimization for molecular orientation search. We will discuss the applicability of the template matching method to Cryo-EM real experimental data.

1Pos021 (1SFP-4) 創薬標的タンパク質の中性子結晶構造解析
(1SFP-4) Neutron crystallographic analysis of drug-target proteins

Takeshi Yokoyama (*Fac. of Pharm. Sci., Univ. of Toyama*)

Neutron crystallography enables the direct visualization of hydrogen/deuterium atoms in proteins and thus is a useful tool for revealing protein-drug interactions in detail. In this decade, we determined the neutron crystal structures of transthyretin (TTR), farnesyl pyrophosphate synthase (FPPS) and bromodomain-containing protein 4 (BRD4). TTR is associated with hereditary ATTR amyloidosis. FPPS and BRD4 are the targets of osteoporosis and anti-cancer drugs, respectively. Based on the neutron structures, we will discuss the molecular stability of TTR associated with amyloidogenesis, the protonation state of the bisphosphonates bound to FPPS and the molecular origin of the binding enthalpy of BRD4 inhibitors.

1Pos022 Crystal structure of human Dishevelled1 PDZ with its inhibitor

Shotaro Yasukochi[1], Nobutaka Numoto[2], Kiminori Hori[1], Natsuko Tenno[1], Takeshi Tenno[1], Nobutoshi Ito[2], Hidekazu Hiroaki[1] ([1]*Grad. Sch. Pharm Sci., Univ. Nagoya*, [2]*Med Res Inst., TMDU*)

PDZ domains mediate protein-protein interactions (PPI) and assemble signaling proteins, thereby regulating various cellular signals. Many non-peptidic PDZ domain inhibitors have been developed, little is known about the structures of PDZ-inhibitor complexes. We reveal the X-ray crystallographic structure of the PDZ domain from the Wnt signaling pathway protein human Dishevelled1 (hDvl1) in complex with and in absence of its inhibitor NPL-3009. Our structures showed that hDvl1 PDZ domain possesses a binding pocket with a unique shape, which is different from previously reported hDvl2 PDZ domain. NPL-3009 bound to this uniquely-shaped pocket and its carboxylate moiety acted as an anchor for specific molecular recognition.

1Pos023* 拡張アンサンブル法によるビタミン D 受容体のアゴニスト/アンタゴニスト活性調節機構の研究
Regulation mechanism of agonistic / antagonistic activities of vitamin D receptor studied by generalized ensemble method

Takafumi Kudo[1], Toru Ekimoto[1], Tsutomu Yamane[1], Mitsunori Ikeguchi[1,2] ([1]*Grad. Sch. Med Life Sci., Yokohama City Univ.*, [2]*Med. Sci. Innov. Hub., Riken*)

Vitamin D receptor (VDR) is one of the nuclear receptors and an important target for drug discovery. To clarify the regulation mechanism of agonistic/antagonistic activities of VDR ligands, molecular dynamics simulations (MD) were performed. Since the crystal structures of agonist and antagonist complexes of VDR were almost identical, it is difficult to discuss the differences of their activities. It is also a problem that it is difficult to observe large conformational changes using nomal MD. Therefore, gREST (Kamiya et al., JCP 2018), which is one of the generalized ensemble methods was utilized. As a result, spontaneous conformational changes of VDR were captured, that made it possible to consider the cause of the differences of agonistic/antagonistic activities.

1Pos024* MARTINI 力場を用いた粗視化シミュレーションによる分子シャペロニン GroEL の ATP に誘起される構造変化の解析
Analysis of the ATP-induced conformational change of the molecular chaperonin GroEL by coarse-grained simulations using the MARTINI

Yuya Yamaura, Tadaomi Furuta, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech*)

The chaperonin GroEL assists protein folding using ATP energy. It is known that ATP binding induces a large structural change from a closed to an open conformation. However, the detailed mechanism of this conformational change is still unclear. To elucidate the mechanism of the ATP-induced conformational change of GroEL, we conducted coarse-grained simulations using the MARTINI force field for the holo (ATP-bound) and the apo (ATP-free) states. As a result, in the holo state, the structural change from a closed to an open conformation was observed with a high probability, whereas such a change was never observed in the apo state. Moreover, it was suggested that changes in the salt bridge networks would contribute to the conformational change of GroEL.

1Pos025* A Theoretically Study of ATP Effect on Solubility of Intrinsically Disordered Protein under Crowded Environment

Hayato Aida[1], Ryuhei Harada[2], Yasuteru Shigeta[2] ([1]*Coll. Bio. Sci., Univ. Tsukuba*, [2]*CCS, Univ. Tsukuba*)

Intrinsically Disordered Protein (IDP) plays an important role in liquid-liquid phase separation (LLPS). RNA-binding protein Fused in sarcoma (FUS) is one of the proteins inducing LLPS because the low-complexity domain of FUS (FUS LC) is IDP. Recently, it was elucidated that ATP concentration can modulate droplets formed by LLPS. In the present study, we performed all-atom MD simulations for the 57 residues of FUS LC (FUS57) under its crowded conditions. As a result, the values of radius of gyration (Rg) in the crowded conditions were larger than those of Rg in the dilute condition, enabling FUS57 to occupy a large space via intermolecular interactions. Finally, we would like to discuss the ATP-FUS57 interaction by performing ATP-included/FUS57-crowded MD simulations.

1Pos026* 脂質膜表面におけるシトクロム P450 還元酵素の誘電アロステリー
Dielectric allostery in cytochrome P450 reductase on the surface of lipid membrane

Mikuru Iijima, Jun Ohnuki, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

Cytochrome P450 reductase (CPR) supplies electrons from NADPH to heme-containing proteins via FAD and FMN, which is considered to be regulated by the open-closed motion between FAD and FMN domains coupled with the redox state. In our previous molecular dynamics study, we showed the coupling between the redox and structural states and found that the dielectric allostery (i.e., a large-scale dielectric response caused by the redox-state change) underlies this coupling. Since CPR is anchored to the lipid membrane in a cell, we here examine the effect of the lipid membrane on the redox-structural state coupling of CPR. We focus on the surface charge of the lipid membrane and discuss the role of the dielectric property of CPR in the electron transfer on the membrane.

1Pos027* TypeIII 中間径フィラメントの細胞表面上への出現機構の解明
Elucidation of recruitment mechanism of type III intermediate filament proteins to cell surface

Beomju Hwang[1], Inu Song[1], Hirohiko Ise[2] ([1]*Grad. Sch. Eng., Kyushu univ.*, [2]*IMCE, Kyushu Univ.*)

Type 3 intermediate filament proteins (3IFs) comprised of vimentin, desmin, GFAP, and peripherin have been reported to express on cell surfaces of various cells. However, the mechanisms of the cell surface expression for these proteins remain unclear. In this study, to elucidate the cell-surface expression mechanisms, we examined whether 3IFs have an affinity to cell membrane by the interaction of 3Ifs with lipid bilayers with surface plasmon resonance (SPR). These results demonstrated that approximately 4-mer of 3IFs had high affinity to lipid bilayers. Moreover, 4-12-mer of 3IFs exposed to the cell surfaces but not the filamentous 3Ifs. We suggest that 4-12-mer of 3IFs depolymerized from the filamentous 3IFs are easy to be recruited to cell membrane.

1Pos028* 量子化学計算による EcoRV のシシルリン酸基ツイスト後のリン酸エステル加水分解反応の研究
DNA Hydrolysis by EcoRV Subsequent to Scissile-Phosphate Twist, Studied by QM/MM Metadynamics Simulation

Itaru Onishi[1], Norio Yoshida[2], Fumio Hirata[3,4], Masayuki Irisa[1] ([1]*Kyushu Inst. of Tech.*, [2]*Kyushu Univ.*, [3]*IMS*, [4]*Toyota Riken*)

A QM/MM metadynamics simulation of *Eco*RV–DNA complex structure was performed with the initial structure having rearranged DNA fragments obtained from the MD trajectory in our previous study. Our results show that a dissociative pathway is the most favorable for DNA hydrolysis by *Eco*RV. *Eco*RV is a type II restriction enzyme, which requires Mg^{2+} for its activity. However, the positions of Mg^{2+} during the DNA hydrolysis by *Eco*RV and mechanism of the reaction are unclear. The potential of mean force surface obtained from metadynamics simulation shows that the activation energy of dissociative pathway is 12kcal/mol, which is 3 kcal/mol lower than that of associative pathway.

1Pos029 Crystal structure of branched-chain polyamine synthase

Eiichi Mizohata[1,2], Masataka Toyoda[1], Ryota Hidese[3], Shinsuke Fujiwara[3] ([1]*Grad. Sch. Eng., Osaka Univ.*, [2]*JST-PRESTO*, [3]*Grad. Sch. Sci. Tech., Kwansei Gakuin Univ.*)

Polyamines are low molecular weight basic substances composed of carbon chain and amino group. The thermophilic bacteria have long-chain or branched-chain polyamines in addition to general polyamines. Branched-chain polyamines are synthesized from spermidine using decarboxylated S-adenosylmethionine as an aminopropyl group donor by branched-chain polyamine synthase (BpsA). A hyperthermophilic archaeon Thermococcus kodakarensis contain only a quaternary amine N4-bis (aminopropyl) spermidine. On the other hand, a thermophilic bacterium Thermus thermophilus has diverse branched-chain polyamines such as tertiary amine N4-aminopropylspermidine. To elucidate the molecular mechanism of polyamine synthesis, X-ray crystallography of two kinds of BpsA was carried out.

1Pos030 Analysis of the complex molecular system composed of GGA, MPR and Ub by using titration SAXS measurement

Yugo Hayashi[1], Natsumi Endo[1], Youichi Yamazaki[1], Sachiko F. Toma[1], Hironari Kamikubo[1,2] ([1]*Div. Mat. Sci., NAIST*, [2]*IMSS KEK*)

The GGAs are a family of clathrin coat adaptor proteins involved in vesicular transport, which is composed of three domains of the VHS, GAT, and GAE domains. GAT acts as a platform for multiple interaction partners, such as MPR, Ub, and so on. Here we studied the regulation mechanism of the interactions between VHS and MPR, GAT and Ub. We revealed the GGA takes a compact form in the absence of the MPR and Ub. The binding with the MPR peptide induced the domain rearrangement of GGA resulting in the release of VHS from GAT. We carried out continuous titration SAXS measurements in the presence or absence of MPR. In the results, we revealed that the Kd of Ub against GAT is influenced by the interaction between VHS and MPR.

1Pos031 Structural analysis of firefly luciferase with MM and QM/MM molecular simulations to clarify the origin of emission color-change factors

Kota Nosaka, Naohisa Wada (*Grad. Sch. Life Sci., Univ.Toyo*)

Red and yellow-green lights are emitted Luciola cruciate bioluminescence in vitro under acidic and basic conditions, respectively. Mutations of its enzyme luciferase (Luc) also produce emission color-changes. In order to investigate factors that cause emission color-changes, we calculated the pK values of some amino acid residues in proximity to the Luc's ligand by molecular simulation, using not only MM, but also QM/MM where the Luc's ligand is only considered within the QM region; meanwhile all amino acid residues of Luc and some water molecules are incorporated into the MM region. Then we compare the results of QM/MM with those of MM to clarify how the effect of QM manifests in the structure of Luc and its mutations.

1Pos032 シアノバクテリア概日時計における KaiC 六量体の構造多様性
Structural Diversity of KaiC Hexamer in Cyanobacterial Circadian Clock

Yoshihiko Furuike[1,2], Atsushi Mukaiyama[1,2], Eiki Yamashita[3], Takao Kondo[4], Shuji Akiyama[1,2] ([1]*Research Center of Integrative Molecular Systems (CIMoS), Institute for Molecular Science (IMS)*, [2]*Department of Functional Molecular Science, SOKENDAI (The Graduate University for Advanced Studies)*, [3]*Institute for Protein Research, Osaka University*, [4]*Graduate School of Science, Nagoya University*)

KaiA, KaiB, and KaiC constitute a core oscillator in the cyanobacterial circadian clock. KaiC is a key enzyme cycling its own ATP/ADP selectivity, phosphorylation states, and quaternary structures in a circadian manner. Despite such diverse states distinguished chemically, the corresponding structural diversity in 3D space has remained poorly characterized. We recently crystallized KaiC in a hexameric form under several conditions and identified multiple unique conformations. Together with biochemical assays of KaiC mutants designed on the bases of the present structures, elasticity, plasticity, and flexibility afforded to KaiC structures will be discussed.

1Pos033 NMR analysis of metal ion-induced conformational changes of α-helical peptides

Ikuko Iizumi[1], Yohei Miyanoiri[2], Toshiki Tanaka[3], Masayuki Oda[1] ([1]*Grad. Sch. Life Environ. Sci., Kyoto Pref. Univ.*, [2]*Inst. Protein Res., Osaka Univ.*, [3]*Nagoya Inst. Technol.*)

We designed simple model peptides which formed three helix-bundle structures induced by metal-ion binding to analyze structural dynamics in protein folding and interaction. In this study, we analyzed the conformational changes of two peptides, HA and HL, whose hydrophobic residues are His/His/Ala and His/His/Leu, respectively. Circular dichroism experiments indicated that the α-helix content of both peptides increased upon the binding of Zn^{2+}. NMR experiments showed that the signals in the presence of Zn^{2+} were dispersed, in contrast to those in the absence of Zn^{2+}. We discuss the site-specific conformational changes of HA and HL analyzed using NMR, and the correlation with the binding thermodynamics analyzed using isothermal titration calorimetry.

1Pos034 シクロスポリン A の CHARMM 力場の開発と膜—水系の分子動力学シミュレーションへの応用
Development of the CHARMM force field for Cyclosporine A and application to molecular dynamics simulations using a membrane-water system

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Cyclosporin A (CsA) is a cyclic peptide formed by 11 residues and used as an immune suppresser. CsA has the permeability to bio-membrane, although its high molecular weight (M. w. ~1200) because of the conformational change between the structures with a hydrophobic surface (closed form) and with a hydrophilic surface (open form). CsA contains unnatural residues (N-methyl amino acid), which contribute to the conformational change of CsA. Here, we developed the CHARMM force field of CsA and performed MD simulations in chloroform solvent. These results were consistent with the conformations of crystal structures and NOE from NMR measurements. Also, we performed MD simulations in the water-membrane system and analyzed the dynamics in the lipid bilayer.

1Pos035 ヒト成人ヘモグロビンの酸素親和性制御に関連した GHz, THz 領域振動の研究
Study on Giga- and Terahertz-frequency Motions Involved with Oxygen Affinity of Human Adult Hemoglobin

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To investigate experimentally a hypothesis proposed by Prof. Yonetani, in which amplitude fluctuations of globin controls oxygen affinity of hemoglobin, we have carried out gigahertz and terahertz spectroscopy for hemoglobins (Hb A) with different oxygen affinities in 0.075 - 1.5 THz using a terahertz-time domain spectrometer and a vector network analyzer. We dissolved Hb A in mixture solvents of water and glycerin to decrease the dielectric constant of water. Difference spectra of absorption in 0.075 - 1.5 THz between deoxy (low affinity) and oxy (high affinity) Hb A showed no spectral structure. Therefore, to reduce more dielectric constant of solvent around Hb A, we have mixed nano-pore silica gel and Hb A, and have preliminarily measured THz spectra of its sample.

1Pos036 Structural basis for the intramolecular signal transduction of oxygen sensor protein FixL from Bradyrhizobium japonicum

Misaki Kamaya[1], Hiroyasu Koteishi[1], Takehiko Tosha[1,2], Seiki Baba[3], Hiroshi Sugimoto[1,2], Yoshitsugu Shiro[1], Hitomi Sawai[1,2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*RIKEN SPring-8*, [3]*SPring-8 / JASRI*)

Rhizobia are responsible for nitrogen fixation that converts nitrogen in the atmosphere into the ammonia. An oxygen sensor system regulates the gene expression of enzymes for nitrogen fixation, because their catalysis requires an anaerobic condition. The FixL from Bradyrhizobium japonicum is a histidine kinase that senses the oxygen tension in the cytoplasm through a heme-containing PAS domain and transfers phosphate from ATP to His residues within histidine kinase domain, and then to the response regulator protein FixJ. In this study, crystal structure and site-directed mutagenesis analysis of FixL suggested that the residues of PAS and coiled-coil region on the dimer interface have a regulatory role in the intra-molecular signal transduction from PAS to kinase domain.

1Pos037 心筋ナトリウムチャネル Nav1.5 と薬剤間の結合自由エネルギー計算
Calculation of the binding free energies between the Nav1.5 sodium channel and drug molecules

Tatsuki Negami[1], Tohru Terada[2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

All drug candidates are tested for cardiotoxicity. Predicting interactions between drugs and cardiac channels, hERG, Nav1.5, and Cav1.2, is important for the proarrhythmia assessment. Previously, we have developed a method to predict the hERG-drug binding affinity based on molecular docking simulations and free energy calculations using the MP-CAFEE method. In this study, we applied this method to the Nav1.5 sodium channel. First, we constructed several structural models based on the different experimental structures of related sodium channels. Then, drugs were docked to the models and the binding free energies were calculated for the top ranked poses. We will discuss the difference in the docking poses and the binding free energies between the Nav1.5 structural models.

1Pos038 DNA 修復にかかわる Hef の天然変性領域の構造と機能
The structure and function of intrinsically disordered region of Hef that is associated with a DNA repair

Takashi Oda[1], Ayako Sekino[1], Ayaka Murakami[1], Rika Oi[1], Maki Yoneyama[1], Noriyuki Kodera[2], Toshio Ando[2], Tsuyoshi Konuma[3], Kenji Sugase[3], Tomotaka Oroguchi[4], Sonoko Ishino[5], Yoshizumi Ishino[5], Mamoru Sato[1] ([1]*Grad. Sch. of Med. Life Sci., Yokohama City Unv.*, [2]*WPI NanoLSI, Kanazawa Univ.*, [3]*Grad.Sch. of Eng., Kyoto Univ.*, [4]*Faclt. Sci. Tech., Keio Univ.*, [5]*Grad. Sch. of Bioresource & Bioenviron. Sci., Kyushu Univ.*)

Hef is an archaeal protein associated with a repair of damaged DNA. Hef has helicase and nuclease domains. Both domains are connected by intrinsically disordered region (IDR) composed of 100 amino acid residues. However, the function of the IDR (Hef IDR) for a DNA repair is still unknown. We have elucidated that Hef IDR interacts with both PCNA and DNA, thus suggesting that it suppresses rapid sliding of PCNA on dsDNA. To investigate the function of Hef IDR for the sliding of PCNA, we carried out high-speed AFM analysis. Biochemical and structural analyses based on SAXS, NMR, and MD indicate that the N-terminal region of Hef IDR forms an α-helix and is required for the interaction with DNA. In this session, we discuss the structure and function of Hef IDR.

1Pos039 T2-like ファージ宿主認識蛋白質と宿主 OmpC の相互作用解析
Structural and functional analysis of phage receptor binding protein and OmpC

Shuji Kanamaru (*Dep. of Life Sci. & Tech., Tokyo Inst. of Tech.*)

T2-like phages have unique receptor binding protein (RBP), gp (gene product) 38, at the distal tip of the long tail fiber. Mutational analysis has been reported that the RBP recognize and identify host bacteria surface LPS and/or outer membrane proteins. From the X-ray structure of phage RBP, gp38, has 5 loops sticking out towards the host. It is most likely that these regions are responsible for host recognition. However, the recognition details are still unknown. Here I will discuss the structural and functional analysis of RBP and target membrane protein OmpC.

1Pos040 ヤナギマツタケ（Agrocybe cylindracea）の Pri3 遺伝子のクローニングと特性解析
Cloning and characterization of the Pri3 gene of the edible mushroom, Agrocybe cylindracea

Chika Abematsu[1], Yamato Kuratani[1], Masashi Shin[1], Makoto Iwata[2], Toshihiko Matsumoto[1], Shoji Ando[1] ([1]*Fac. Biotech. Life Sci., Sojo Univ.*, [2]*IMB*)

Molecular mechanisms of fruiting body formation of edible mushrooms are poorly understood. The Pri3 gene has been reported to be specifically expressed in the primordia of the Agrocybe aegerita complex, although physiological function of the translated product PRI3 is still unknown. We cloned the Pri3 cDNA from Agr. cylindracea cultivated in Fukuoka Prefecture, and characterized its expression pattern in the mushroom by Northern blotting and in situ hybridization. The localization of the PRI3 protein in the mushroom was also determined by immunohistochemistry. The results obtained suggest that the Pri3 gene is expressed specifically in the pileus during fruiting body maturation, but not in the primordia.

1Pos041 ヒトヘアケラチン K85 の遺伝子導入細胞における機能特性と外胚葉形成不全症の原因となる変異の影響
Functional characteristics of human hair keratin K85 in transfected cells and the effects of mutations causative of ectodermal dysplasia

Masaki Yamamoto[1], Yasuko Sakamoto[1], Yuko Honda[2], Kenzo Koike[3], Hideaki Nakamura[4], Toshihiko Matsumoto[1], Shoji Ando[1] ([1]*Fac. Biotech. Life Sci., Sojo Univ.*, [2]*Fac. Med., Saga Univ.*, [3]*Kao corp.*, [4]*Fac. Phar., Sojo Univ.*)

Human type I hair keratin 35(K35) and type II hair keratin 85(K85) are the first pair expressed in the early differentiation of hair-forming cells. Two kinds of mutations in the K85 gene have been identified in the families with ectodermal dysplasia of hair and nail type. To study the filament forming ability of the K85-K35 pair with or without the disease-related mutations, the K85 genes were transiently co-expressed with the K35 gene in human cultured cells. The structures and localization of the filaments produced by the hair keratin pairs varied depending on the types of endogenous intermediate filaments in the cells, location of the mutations in the K85 gene, and elapsed time after transfection. The structure-activity relationship of K85 will be discussed.

1Pos042 巨大ヘモグロビンのアロステリック中間体の時分割構造解析
Time-resolved structure analysis of allosteric intermediate of the giant hemoglobin

Nobutaka Numoto[1], Yoshihiro Fukumori[2], Kunio Miki[3], Nobutoshi Ito[1] ([1]*Med. Res. Inst., Tokyo Med. Dent. Univ. (TMDU)*, [2]*College Sci. Eng., Kanazawa Univ.*, [3]*Grad. Sch. Sci., Kyoto Univ.*)

Allosteric oxygen-binding mechanism of hemoglobin (Hb) has been widely discussed whereas the structure of intermediate form between the oxy and deoxy states without any artificial modification of the Hb molecule is still unclear. The oxygen molecules bound to the extracellular giant Hb (400 kDa) of a tubeworm, *Oligobrachia mashikoi*, were gradually dissociated in a crystalline state, and we obtained four distinct oxy-deoxy intermediate forms, which are confirmed by the microspectrophotometric analyses. The crystal structures of these intermediates provide coarse snapshots of allosteric intermediate of the giant Hb and details of the ternary and quaternary structural changes, as well as the oxygen dissociation ratio in each subunit.

1Pos043* 分子動力学法による RvSAHS1 の構造安定性
Structural stability of RvSAHS1 by MD simulations

Kazuhisa Miyazawa[1,2,3], Satoru Itoh[1,2,3], Hisashi Okumura[1,2,3] ([1]*SOKENDAI*, [2]*IMS*, [3]*ExCELLS*)

Tardigrades survive some extreme environmental conditions by entering cryptobiosis state. SAHS protein family, including RvSAHS1, is thought to be related to the cryptobiosis. While the crystal structure of RvSAHS1 was determined recently, the structure comprises some crystalline solvents, and does not include intrinsically disordered protein (IDP) region which is a feature of RvSAHS1. We investigated the properties of RvSAHS1, which includes no crystalline solvents and has IDP region, by molecular dynamics simulations. We revealed three properties, the flexibility of entrance region, the fluctuation of IDP region and the contact property of the IDP region. Based on the result, we hypothesized the role of RvSAHS1 on the cryptobiosis.

1Pos044* Rheo-NMR 法によるスーパーオキシドジスムターゼ 1 の動的なアミロイド形成機構の解析
Dynamic Analysis of Amyloid Formation of Superoxide Dismutase 1 Using Rheo-NMR Spectroscopy

Naoto Iwakawa[1], Daichi Morimoto[1], Erik Walinda[2], Masahiro Shirakawa[1], Kenji Sugase[1] ([1]*Grad. Sch. Eng., Kyoto Univ.*, [2]*Grad. Sch. Med., Kyoto Univ.*)

Superoxide dismutase 1 (SOD1) is an important protein that functions as a radical scavenger in the cytosol, whereas the amyloid formation of SOD1 leads to the onset of amyotrophic lateral sclerosis (ALS). Elucidation of the mechanism by which SOD1 forms amyloid would help the understanding of the onset of ALS. We use Rheo-NMR, which enables in-situ observation of amyloid processes at the atomic level. We detected increases in ^{15}N transverse relaxation rates of residues on the specific surface under shear flow, suggesting the native state interacts with intermediate species. This result provides atomic-level information on amyloid formation, which cannot be obtained by other methods. We plan to analyze the interaction in more detail by using NMR relaxation experiments.

1Pos045* 統計力学モデルの拡張によるタンパク質のフォールディング反応機構の予測
Prediction of protein folding mechanisms by an extended statistical mechanical model

Koji Ooka[1], Munehito Arai[1,2] ([1]*Dept. Phys., Univ. Tokyo*, [2]*Dept. Life Sci., Univ. Tokyo*)

Wako-Saitô-Muñoz-Eaton (WSME) model is a coarse-grained statistical mechanical model and has succeeded in explaining experimentally observed folding mechanisms of small proteins. However, the model cannot be applied to large, multi-domain proteins. To overcome this problem, here we extended the WSME model by introducing non-local interaction terms in the Hamiltonian and calculated free energy landscapes and folding processes of several multi-domain proteins. Using our extended WSME model, we succeeded in theoretically predicting the folding mechanisms of lysozyme and α-lactalbumin, which are consistent with experimental results. We are now working on developing a general method for applying our extended model to various multi-domain proteins.

1Pos046 低波数基準振動で特徴づけられた多量体蛋白質の動的性質
Dynamic properties of oligomeric proteins characterized by low-frequency normal modes

Hiroshi Wako[2], **Shigeru Endo**[1] ([1]*Dept. of Phys., Sch. of Sci., Kitasato Univ.*, [2]*Sch. of Social Sci., Waseda Univ.*)

Dynamics of oligomeric proteins were studied by elastic network model-based normal mode analysis to characterize their large-scale concerted motions. The rigid-body motions of individual subunits were characterized by the radial, tangential, and axial components in a cylindrical coordinate system. The motions of atoms on the inter-subunit interfaces were characterized by the opposing motions of intra-subunit deformation and rigid-body motion of the subunit. A dynamic protein structure network was defined in each mode, and the centrality measure, betweenness, was calculated for each residue. The results indicate that the residues with high betweenness, which are seemed to be more conservative, play an important role in oligomer dynamics.

1Pos047 粗視化 Go モデルを用いた GA・GB ドメイン関連タンパク質のフォールディング機構の相違・共通性の予測
Prediction of differences and commonality in folding mechanisms of GA / GB domain related proteins using coarse-grained Go model

Shoya Hamaue (*Dept. of Bioinfo., Col. of Life sci., Ritsumeikan Univ.*)

The protein-folding is a long-standing problem and there have been a lot of experimental and theoretical studies so far. Based on some of them, it is known that proteins showing the amino acid sequence identity of 30% or more exhibit similar 3D-structures. However, despite showing more than 80% sequence identity, there are several proteins with completely different structures. In this study, we treat artificial proteins with 3 α structure (GA) and 4 β + α structure (GB) derived from albumin binding domain and IgG binding domain, respectively. In order to clarify how such proteins fold into different structures, we conduct simulations based on a coarse grained Go model. As a result, we reveal the differences and commonality in the folding pathway of each protein.

1Pos048 Mechanism of the spontaneous elongation of the fibroin nanofiber involved in spider silk

Takuya Sawai[1], Kiichi Hayashi[1], Yugo Hayashi[1], Takehiro Sato[2], Yoichi Yamazaki[1], Sachiko Toma-Fukai[1], Hironari Kamikubo[1,3] ([1]*Div. Mat. Sci., NAIST*, [2]*Spiber Inc.*, [3]*IMSS KEK*)

Superior mechanical properties of spider silk enable next-generation materials. Fibroin proteins, significant components of the spider silk, consist of repetitive Ala and Gly-rich region, which form the crystalline domains and the amorphous matrix, respectively. Multi-hierarchical architecture was found in the spider silk as well. Recently, we discovered that fibroin proteins spontaneously form the nano-fibrillar structures similar to those in spider silk. In this study, we identified precursors indispensable for the elongation of nanofibers. SAXS and fluorescence measurements revealed that the pre-nanofiber is an oligomeric form of fibroin proteins. From these results, it can be proposed that the nano-fiber formation is composed of nucleation and elongation processes.

1Pos049 天然様階層構造を有する人工クモ糸材料の再構成
Reconstitution of artificial materials of spider silk accompanied by native-like hierarchical structure

Satoru Onishi[1], Yuki Nakatani[1], Yugo Hayashi[1], Takehiro Sato[2], Yoichi Yamazaki[1], Sachiko Toma-Fukai[1], Hironari Kamikubo[1,3] ([1]*Div. Mat. Sci., NAIST*, [2]*Spiber Inc.*, [3]*IMSS KEK*)

Spider silk is expected as high-performance material because of its toughness and heat-resisting property. We previously revealed that fibroin, a significant component of spider silk, spontaneously forms nanofiber in vitro, which appears to be those found in native spider silk. To artificially reconstruct spider silk, we attempted to reproduce multi-hierarchical structure utilizing the fibroin nanofibers. First, hydrogel was prepared with the fibroin nanofibers, and dehydrated under anisotropic tensile stress. The wide angle X-ray diffraction images from the dehydrate hydrogel exhibited anisotropic pattern, suggesting that the dried hydrogel possesses aligned poly-Ala crystal. These structural characteristics are similar to natural spider silk.

1Pos050* ヒト SOD1 と Zn 欠損 SOD との比較による亜鉛と静電ポテンシャルループの役割に関する研究
Investigation on role of zinc atom and electrostatic loop by comparing human SOD1 with Zn-deficient SOD

Natsumi Koyama[1], Masami Lintuluoto[1], Juha Lintuluoto[2] ([1]*Grad. Sch. Life and Environ. sci., Kyoto pref. univ.*, [2]*Grad. sch. eng., Kyoto univ.*)

SOD1 includes Cu and Zn atoms in its reaction site. The previous studies have reported that Cu is the primary reaction site, while Zn plays an important role for decreasing the reduction potential of Cu and stabilizing the conformation. Several point mutations of SOD1 have been reported to induce the aggregation due to Zn deficiency and to cause amyotrophic lateral sclerosis. Although SOD from Mycobacterium tuberculosis (MtSOD) has only one Cu and no Zn, the conformation around Cu is similar to that of SOD1 and the activity is likewise maintained as other Cu, Zn-SODs. The remarkable difference between SOD1 and MtSOD is the length of the electrostatic loop located above the reaction site. In this study, we focus on the zinc role, and electrostatic loop in SOD1 and MtSOD.

1Pos051 点変異導入による CD44 のヒアルロン酸結合のアロステリック制御
Allosteric regulation of hyaluronan binding on CD44 by point mutation

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CD44 plays an important role on the cell rolling via hyaluronan (HA) binding. The conformation of C-terminal on HA-binding domain (HABD), which is connected to the membrane, has been reported to change from a low affinity (ordered, O) form to a high affinity (partially disordered, PD) form under fluid shear stress. We have studied the HA binding on CD44 HABD by using molecular dynamics (MD) simulation. Two conformations of HABD, O and PD forms showed different binding manner and binding affinity. In the present study, we investigated the influence of point mutation on the partially disordered region on the conformation of HABD and the HA binding affinity.

1Pos052 アミロイド β42 を用いた天然変性タンパク質の分子シールド効果の評価
The evaluation of molecular shield effect of intrinsically disordered proteins using amyloid beta 1-42

Koki Ikeda, Yoshiki Shigemitsu, Natsuko Tenno, Takeshi Tenno, Hidekazu Hiroaki (*Grad. Sch. Pharm Sci., Univ. Nagoya*)

In our previous study, we showed that intrinsically disordered proteins (IDPs) have a cryoprotective effect toward enzymes and proteins in a sequence-independent manner. We hypothesized the effect is due to molecular shield, a concept that reduces the stochastic direct collision of protein molecules to suppress aggregation. We further expand the hypothesis from protection of aggregation during freeze-thaw process to inhibition of amyloid fibril formation. In this study, we used amyloid beta 1-42 (Aβ42) as a model protein. We evaluated the molecular shield effect of IDPs by Thioflavin T fluorescence assay during Aβ42 fibrillization at 37°C. All human genome derived IDPs examined showed inhibition of Aβ42 fibrillation at lower concentrations compared to PEG or Dextran.

1Pos053 X線小角散乱を用いた大腸菌フェリチンの鉄コアの形成に関する研究
The iron core formation of E. coli ferritin studied by small angle X-ray scattering

Takumi Kuwata, Daisuke Sato, Kazuo Fujiwara, Masamichi Ikeguchi (*Department of Bioinformatics, Soka University*)

Ferritin (Ftn) is a spherical protein composed of identical 24 subunits. In vivo, Ftn oxidises Fe2+ into Fe3+ and makes an iron core of 8 nm diameter in its cavity. However, the detailed mechanism of iron core formation remains to be clarified. In this study, we investigated the shape of the iron core of Ftn from Escherichia coli (EcFtnA) by small-angle X-ray scattering under the conditions where the electron density of solvent matches that of apo EcFtnA. The matching point was found to be 53% (w/w) sucrose concentration. Under such conditions, we observed the scattering curve from the core of 500-4500 iron atoms. The gyration radius at maximun iron loading was found to be around 30Å which was consistent with the inner diameter of Ftn shell.

1Pos054 柔らかいドライブシャフトを持つ F_1-ATPase のトルク伝達/発生
Torque transmission/generation of F_1-ATPase with a soft driveshaft

Shou Furuike[1], Naoki Soga[2], Yasushi Maki[1], Hideji Yoshida[1] ([1]*Phys., Osaka Med. Coll.*, [2]*Grad. Sch. Eng., Univ. Tokyo*)

F_1-ATPase ($\alpha_3\beta_3\gamma$) is an ATP-driven rotary molecular motor. The shaft of rotor (γ), an antiparallel α-helical coiled coil of the N-ter and C-ter of the γ subunit, is set and fitted in the central cavity of the cylinder-like stator ($\alpha_3\beta_3$-ring). We constructed F_1 mutants which have soft driveshafts. For example, the C-ter α-helix of γ were cut at the middle (the portion would not interact with the $\alpha_3\beta_3$-ring), and joined by “helix-breaker” amino-acids. Because the amid-acids form freely jointed chain, an elasticity of the driveshaft should become weaker. The rotation rates of the mutant without drag were almost same of wild type. In contrast, the torque of the mutant fell below ~3/4 of wild type, indicating the soft driveshaft cannot transmit/generate full torque.

1Pos055* (1SDP-3) Biophysical analysis of alpha-synuclein oligomers by microchip electrophoresis

William E. Arter[1,2], Catherine K. Xu[1], Georg Krainer[1], Christopher M. Dobson[1], Tuomas P. J. Knowles[1,2] ([1]*Centre for Misfolding Disease, Department of Chemistry, University of Cambridge*, [2]*Cavendish Laboratory, Department of Physics, University of Cambridge*)

Alpha-synuclein is a protein strongly implicated in neurodegenerative disease, it is known to misfold and self-assemble into insoluble, fibrillar deposits in the brains of Alzheimer’s patients. Oligomeric aggregates of synuclein are transient, intermediate and toxic species in this process. Studying them remains a challenge for traditional biophysical techniques due to their high degree of heterogeneity, short lifetime and low abundance. Here, we present microfluidic electrophoresis as a novel technique for the biophysical analysis of oligomeric synuclein. Our method enables the highly sensitive study of transient, heterogenous oligomers in the solution-phase, and provides a platform for further examination of protein complexes and protein-protein interactions.

1Pos056* タンパク質内部の構造変化をプローブするためのアスパラギン酸マッピングと赤外分光解析
Mapping of aspartic acids to probe protein structural changes by FTIR spectroscopy

Masanori Hashimoto, Kota Katayama, Manish Singh, Yuji Furutani, Hideki Kandori (*Grad. Sch. Eng., Nagoya Inst. Tech.*)

Stimulus-induced difference FTIR spectroscopy is a powerful method to probe protein structural changes, but detected vibrations have to be assigned by isotope-labeling. On the other hand, carboxylic C=O stretch (1800-1700 cm^{-1}) is well isolated from other vibrations, and if there are no spectral changes for intact protein in this frequency region, introduction of carboxylate by mutation may be a good tool to probe local protein structural changes. This method can also monitor hydrophobicity of the local environment, as the pKa of carboxylates are 4~5 in aqueous solution. A trial of aspartate mapping will be presented for a heliorhodopsin, where light-induced difference FTIR spectra were measured by the ATR technique at room temperature (pH 7).

1Pos057* バイオ医薬品中の蛋白質凝集体の定量手法の確立
Establishment of quantification methods for protein aggregates in biopharmaceuticals

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Several methods for quantification of protein aggregates have been developed and we found quantitative laser diffraction (qLD) is effective. However, different detection principles can yield inconsistent results. This study aimed to compare particle size distributions and concentrations of protein aggregates using orthogonal methods. Protein aggregates were generated by stirring an immunoglobulin solution. Serial dilutions of aggregates stock were analyzed resonant mass measurement (RMM), flow imaging (FI), and qLD. Both particle size distributions and concentrations were in good agreement between RMM and qLD (0.5 to 2 μm) and between FI and qLD (2 to 20 μm). Thus, qLD enables covering of the overlapping analytical ranges of RMM and FI.

1Pos058 (1SDP-6) 新規に開発した高濃度タンパク質のためのネガティブ染色電子顕微鏡法
(1SDP-6) A newly developed negative stain EM method for protein complexes at high protein concentration

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The negative stain EM method is a powerful screening tool to evaluate protein quality prior to cryo-EM. However, the typical protein concentration for negative staining EM is 100 times lower than that for cryo-EM. Proteins weakly associated with a protein complex (e.g. with a Kd in the millimolar range) may fall off by dilution for negative stain EM. Here we have developed a novel negative stain EM method for high concentration protein complexes in order to rapidly visualize a weakly associated protein on a protein complex. We applied this method to glutamate dehydrogenase (GDH), which is known to form a linear polymer only under high concentration. This method has successfully visualized the high-concentration GDH structure. We also observed such structure by cryo-EM.

1Pos059 模倣細胞内分子混雑環境及び糖ガラス中の特定のタンパク質及び脂質膜の構造の中性子散乱による研究
Study of protein or membrane structure in mimicking intra-cell molecular-crowding environment and in sugar-glass using neutron scattering

Mitsuhiro Hirai[1], Satoshi Ajito[1], Shigeki Arai[2], Shinichi Takata[3], Hiroki Iwase[4] ([1]*Graduate School of Science and Technology, Gunma University*, [2]*National Institute for Quantum and Radiological Science and Technology*, [3]*J-PARC Center, Japan Atomic Energy Agency*, [4]*Comprehensive Research Organization for Science and Society*)

The interior of living cells is a molecular-crowding environment. The nature of molecular-crowding is important for an understanding of the elaborate biological reactions and homeostasis occurring therein because the equilibrium states of biological macromolecules are affected by molecular-crowding. However, knowledge about crowding effects is still insufficient due to a lack of relevant experimental studies. Recently, we have found that a new neutron scattering technique is available to directly observe the structure of a specific protein or membrane that exists in small amounts under the presence of large quantities of cell-debris (crowder) [1]. We will also discuss a protein structure embedded in sugar-glass. [1] M. Hirai et al., J. Phys. Chem. B. 2019, 123, 3189.

1Pos060 TRPV1 分子内部の回転動態の決定
Agonist- and antagonist-induced rotational motion of TRPV1 channel

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TRPV1 is a cation channel that responds to various signals including capsaicin, heat, and low pH conditions. Cryo-EM revealed various conformations of TRPV1, supporting its rotational motion in response to ligand binding. However, only limited experimental evidence supports this fact. To understand the dynamics of TRPV1, we adopted the Diffracted X-ray Tracking (DXT) technique. In DXT, the individual protein was labeled with a gold nanocrystal and its intramolecular movement was investigated by tracking the diffraction spot. Here we observed molecular fluctuations of the TRPV1 proteins in tilting and rotational directions, both by capsaicin and AMG9810. The results were further compared to the heat and low pH evoked TRPV1 activation, together with mutation experiments.

1Pos061 レプリカ部分置換法の開発とタンパク質への応用
Development of replica sub-permutation method and its application to mini-protein

Masataka Yamauchi[1,2,3], Hisashi Okumura[1,2,3] ([1]*SOKENDAI*, [2]*ExCELLS*, [3]*IMS*)

We propose a replica sub-permutation method for molecular dynamics and Monte Carlo simulations, which is an improvement of replica-exchange and replica-permutation methods. The replica sub-permutation method introduces a new permutation algorithm referred to as “sub-permutation” to perform temperature and pressure parameter permutation. This method succeeds in reducing the number of combinations between replicas and parameters without the loss of sampling efficiency. We applied this method to the beta-hairpin mini-protein, chignolin, to compare the sampling efficiency with replica-exchange and replica-permutation methods. We show that the replica sub-permutation method is the most efficient method both in the parameter and conformational spaces.

1Pos062 凝縮系の不均一動力学(dynamical disorder)の分子論開拓: タンパク質の構造遷移・揺らぎ階層性
Theoretical investigations on microscopic heterogeneity and hierarchy in transitions and fluctuations of protein conformations

Yoshihiro Matsumura[1], Shinji Saito[1,2] ([1]*IMS*, [2]*SOKENDAI*)

Heterogeneous dynamics or kinetics (dynamical disorder) in protein functions is an important factor. However, knowledge on the atomic and molecular pictures and origins is still very limited. In this study, a series of theoretical methods are developed based on the framework for stochastic process to analyze time-series data of the system by molecular dynamics (MD) simulations and so on. The methods are applied to clarify the roles of dynamical disorder at the microscopic level from single-molecule kinetics perspective, for the molecular theory. As a typical example, ultra-long MD time-series data of BPTI protein dynamics are analyzed to reveal the hierarchy in transitions and fluctuations of conformations, the microscopic pictures, and origins of heterogeneity.

1Pos063 Lytic polysaccharide monooxygenase の生化学および 1 分子解析
Biochemical and single-molecule analyses of lytic polysaccharide monooxygenase

Siti Mastura Zakaria[1,2], Akihiko Nakamura[1,3], Yasuko Okuni[1], Mayuko Yamamoto[1], Akasit Visootsat[1,3], Jun Ando[1,3], **Ryota Iino**[1,3] ([1]*IMS, NINS*, [2]*Univ. of Malaya*, [3]*SOKENDAI*)

Chitin is a highly recalcitrant crystalline polysaccharide composed of *N*-acetyl-D-glucosamine. Lytic polysaccharide monooxygenase (LPMO) plays important role in enzymatic process of the crystalline chitin. In this study, we conducted detailed analyses of chitin degradation activity and single-molecule binding/dissociation kinetics of LPMO from *Serratia marcescens* (SmAA10A). Our results indicate that presence of the co-substrate hydrogen peroxide facilitates binding of SmAA10A to the chitin, and activated SmAA10A stays on the chitin surface for several seconds for degradation. Furthermore, chitin degradation by SmAA10A is not processive, and SmAA10A dissociates from chitin after single catalysis.

1Pos064 Relationship between loop geometry and register shift in parallel beta-sheet proteins

Ryuichiro Ueda, George Chikenji (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

De novo protein design, identifying amino acid sequences that fold into an aimed structure on physical principles, has been significant advanced in recent years. Despite the advances, it is still difficult to select "designable" backbone scaffold for a blueprint. In beta-sheet proteins, the blueprint requires specifying three parameters: length of secondary structures, loop geometry, and register shift. There are, however, no reasonable criteria to specify optimal register shift. Here we propose practical criteria for parallel beta-sheet proteins. The criteria were drived from database analysis and simulation.

1Pos065 2 アミノ酸同時変異戦略を用いた SBDD による超高親和性抗体の創製
Structure-based discovery of the antibodies with sub-picomolar affinity using the double amino acid mutation strategy

Shuntaro Chiba[1], Masateru Ohta[1], Aki Tanabe[2], Makoto Nakakido[2], Kouhei Tsumoto[2,3], Yasushi Okuno[1,4] ([1]*MIH, RIKEN*, [2]*Sch. Eng., Univ. Tokyo*, [3]*Inst. Med. Sci., Univ. Tokyo*, [4]*Grad. Sch. Med., Kyoto Univ.*)

In the design of antibody, the affinity is one of the critical points to be improved. A typical strategy for this begins with single-point mutations, followed by a combination of good or not-too-bad mutants. In this study, a new strategy "double-point mutations" was taken to find residue pairs having good interactions with antigen and also between the mutated residues. The antibody-antigen pair with 53-pmol/L affinity was chosen as a target for the strategy. Tens of thousands of 3D models of single- and double-point mutants were computationally generated. Using structure- and interaction-based selection, ten mutants were selected. The SPR experiments showed the affinity improvement in four mutants, two of which exhibited over 30-fold and 200-fold improvements.

1Pos066 転写コアクチベータ CBP の KIX ドメインと転写因子間の相互作用を阻害するペプチドの合理的設計
Rational design of peptides that inhibit interactions between the KIX domain of CBP and transcription factors

Nao Sato[1], Shunji Suetaka[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

The KIX domain of a transcriptional coactivator CREB-binding protein (CBP) binds many kinds of transcription factors, which may cause leukemia and various viral diseases, including AIDS. Inhibiting the interactions between them may help develop drugs for such diseases. Here, we design peptides that strongly bind KIX at the binding site of transcription factors. As a template for peptide design, we use the transactivation domain of the mixed lineage leukemia transcription factor (MLL), which is important in hematogenesis and early embryogenesis. Using the protein design software Rosetta, we design peptides that binds KIX more tightly than MLL. We will report our progress at the meeting.

1Pos067 分子動力学シミュレーションを用いた CDR-Grafting による合成単ドメイン抗体の構造的変化の解析
Structural effects of CDR grafting of synthetic single domain antibodies investigated by molecular dynamics simulations

Seisho Kinoshita[1], Chinatsu Mori[2], Makoto Nakakido[1,2], Daisuke Kuroda[1,2,3], Jose Caaveiro[4], Kouhei Tsumoto[1,2,3,5] ([1]*Dept. of Bioeng., Sch. of Eng., Univ. of Tokyo*, [2]*Dept. of Chem. Biotech., Sch. of Eng., Univ. of Tokyo*, [3]*Med. Dev. Dev. Reg. Res. Center, Sch. of Eng., Univ. of Tokyo*, [4]*Grad. Sch. of Pharm. Sci., Kyushu Univ.*, [5]*Inst. of Med. Sci., Univ. of Tokyo*)

Camel single domain antibody fragments (VHHs) are useful tools in biotechnological and medical applications. To further improve their applicability, optimizations based on the understanding of molecular mechanisms and physicochemical properties are required. Herein we have exchanged the complementarity determining regions (CDR) between different scaffolds and performed physicochemical analyses, revealing that synthetic VHHs exhibited various thermal stabilities and affinities, even with the same CDRs, depending on the scaffold. We hypothesized these differences were caused by structural fluctuations. To examine this idea at the atomic level we employed molecular dynamics simulations. In light of these data, we discuss possible mechanisms explaining these differences.

1Pos068 アレルギー性喘息に関わるインターロイキン 33 の阻害剤開発に向けて
Toward the development of an inhibitor of interleukin-33 responsible for allergic asthma

Mio Sano[1], Yoshiki Oka[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

Interleukin 33 (IL-33) induces allergic diseases, especially allergic asthma, upon binding to the ST2 receptor on the surface of type 2 innate lymphoid cells. Thus, an inhibitor of IL-33-ST2 interaction is expected to be a therapeutic agent for allergic asthma. Here, using three Ig-like domains (D1-D3) in the ectodomain of ST2 as templates, we attempted to rationally design the proteins that inhibit IL-33 binding with ST2. Among various ST2 fragments constructed, only the D1 fragment of ST2 was easily purified, although its interaction with IL-33 measured by isothermal titration calorimetry was weak. Using protein design software Rosetta, we are now designing the mutants of the D1 and other ST2 fragments that can bind tightly to ST2 and are easy to be purified.

1Pos069 Domain-specific monitoring of higher-order structure of therapeutic IgG on the basis of molecular recognition of artificial proteins

Hideki Watanabe[1], Naoko Hayashida[2], Shinya Honda[1,2] ([1]*BMRI, AIST*, [2]*Grad. Sci. of Fro., Univ. of Tokyo*)

For biosensing-based quality control of therapeutic IgG, we have previously generated an artificial protein termed AF.2A1 that specifically recognizes non-native structure of the Fc region of IgG. This analytical technique can be further advanced by expanding the variety of analytical probes. In the current study, we performed phage display selection of novel artificial proteins targeting non-native structure of the Fab region. An artificial protein termed AF.ab920 specifically recognized higher-order structural change of the Fab region; AF.ab920 bound to non-natively folded Fab region, but not to natively folded Fab or non-natively folded Fc region. These analytical probes enable domain-specific monitoring of higher-order-structure of therapeutic IgG.

1Pos070 主鎖環状化が顆粒球コロニー刺激因子に与える会合体抑制効果
An effect of resistance to self-association of backbone circularization on granulocyte-colony stimulating factor

Risa Shibuya[1], Takamitsu Miyafusa[2], Shinya Honda[1,2] ([1]*Grad. Sch. of Fro., Univ. of Tokyo*, [2]*BMRI, AIST*)

Granulocyte-colony stimulating factor (G-CSF) is a cytokine which is widely used as biopharmaceuticals in the treatment of leukopenia. However, G-CSF has the problem of highly self-association at physiological and formulation pH. Protein drugs containing nonnative structures are expected to be less functional than native proteins, or immunogenic at the administration stage. Therefore, reducing the self-association of G-CSF is important for manufacturing effective and safe therapeutics. Previously, we showed that G-CSF is amenable to conformational stabilization by the backbone circularization. Here, we investigated whether backbone circularization suppress self-association of G-CSF by examining the difference of self-association between linear and circularized G-CSF.

1Pos071 Function of N-terminal acetylation of fish hemoglobin α-subunit

Satoru Unzai, Antonio Tsunesige (*Hosei University, Department of Frontier Bioscience*)

Teleost fish commonly exhibit remarkable hemoglobin (Hb) multiplicity with marked differences in the oxygen binding properties of individual Hbs and in their sensitivities to allosteric effectors. The differences may serve to adapt oxygen transport to environmental variations and metabolic requirements. On the other hand, acetylation of N-terminal amino groups is very common in fish hemoglobin α-subunit, but not in β-subunit. What is the specific function of this modification in fish Hb? Trout Hb IV, one component of the rainbow trout Hb system, shows a very strong pH effect (Root effect) and remarkable sensitivity to organic phosphates. The relationship between the acetylation modification and oxygen binding properties of the trout Hb IV is examined in this study.

1Pos072 Functional roles of conserved residues near the active site of nitric oxide reductase based on the structural analysis

Takehiko Tosha[1], Raika Yamagiwa[2], Takuya Kurahashi[2], Hiroshi Sugimoto[1], Yoshitsugu Shiro[2] ([1]*RIKEN SPring-8*, [2]*Univ. of Hyogo*)

Membrane-integrated nitric oxide reductase (NOR) catalyzes the decomposition of cytotoxic nitric oxide (NO) in microbial anaerobic respiration called denitrification at the heme/non-heme iron binuclear center through the reaction, 2NO + 2H+ + 2e- => N2O + H2O. To understand the mechanism of the NOR-catalyzed reaction based on the mutational analysis, we established a homologous expression system for NOR from Pseudomonas aeruginosa. In addition, we determined the crystallization condition for the recombinant NOR with lipidic cubic phase method to interpret the mutational effects on the function at the structural level. Using newly developed systems, we explored the roles of several conserved residues located near the binuclear active center in the function of NOR.

1Pos073 2—状態アロステリーモデルの再検討ー機能的階層の有無
INSIGHT INTO THE TWO-STATE ALLOSTERIC MODEL - ON THE EXISTENCE OF HIERARCHIES

Antonio Tsuneshige, Satoru Unzai (*Hosei Univ. Frontier Bioscience*)

We report about dramatic changes in allosteric function of Hb that are caused by specific chemical alterations of the α1β1, an interface that has been considered inert. The modified α1β1 interface Hbs exhibited characteristics of extreme T-state species, i.e., pronounced low affinity for oxygen and almost absence of cooperativity, which cannot be explained by the classical TwoState MWC model. Current experiments are directed to prove whether these effects are solely present in the tetrameric structure, or are characteristics of a composite of sub-hierarchical conformations, as is the case of a dimer. If so, a modification of the Two-State Allosteric Model that comprises these neglected conformations must be taken into consideration.

1Pos074* (1SCA-2) 脂質分子の混み合い効果による膜貫通タンパク質結晶化の検討
(1SCA-2) Crystallization of transmembrane protein driven by molecular crowding effect of lipids: Theoretical estimation by using a simple model

Keiju Suda[1], Ayumi Suematsu[2], Ryo Akiyama[1] ([1]*Kyushu University, Sci.*, [2]*Kyushu Sangyo University, Science and Engineering*)

We studied driving force of transmembrane protein crystallization by using a two-dimensional binary hard disk system, namely a mixture of lipids and proteins. Wild type bacteriorhodopsin (bR) trimers crystallize in a lipid bilayer. On the other hand, some mutant bRs do not construct trimers, and the critical concentration(CC) of crystallization is 10.2 times higher than wild type bRs. The free energy curves are obtained based on the free volume theory and the scaled particle theory and phase diagrams are drawn. The phase diagrams for trimer- and monomer- lipid systems are compared and the ratio between CCs (CCR) are obtained. Calculated CCR agrees with the experimental one. This result suggests that the depletion force plays an important role in the crystallization.

1Pos075* アミロイド前駆体タンパク質とβ切断酵素の膜貫通部位の生体膜中での相互作用
Interaction between amyloid precursor protein and beta-secretase in the bio-membrane

Kaori Yanagino, Naoyuki Miyashita (*BOST KINDAI*)

Alzheimer's disease is known as one of dementia, and millions patients have existed in Japan. Amyloid Precursor Protein (APP) is cleaved by beta-secretase in lipid raft membrane and amyloid beta peptide is produced in the early stage of Alzheimer's disease. The amyloid beta peptides are aggregated each other, and construct the senile plaque. Thus, interactions between APP and beta-secretase are very important of the proceeding of the disease. We, therefore, performed the structure prediction of the transmembrane domain (TM) of beta-secretase using Replica-exchange molecular dynamics simulations (REMD). We also performed the coarse grained model Molecular dynamics simulations of the TM domain of beta-secretase and APP in the lipid raft environments.

1Pos076* 多次元固体 NMR による細胞膜中のヘリオロドプシンの構造解析
Multidimensional solid-state NMR study of heliorhodopsin in a lipid environment

Shibuki Suzuki[1], Toshio Nagashima[2], Rina Kaneko[1], Takashi Okitsu[3], Akimori Wada[3], Naohiro Kobayashi[2], Toshio Yamazaki[2], Rei Abe-Yoshizumi[4], Keiichi Inoue[5], Hideki Kandori[4], Izuru Kawamura[1] ([1]*Grad. Sch. Eng. Sci., Yokohama National Univ.*, [2]*RIKEN RSC*, [3]*Kobe Pharmaceutical Univ.*, [4]*Nagoya Inst. Tech.*, [5]*Univ. Tokyo*)

Heliorhodopsin is a transmembrane protein with a retinal chromophore that constitutes a new rhodopsin family different from type-1 and type-2 rhodopsin in views of sequence homology, membrane protein topology and function [1]. We performed multidimensional solid-state NMR experiments of a HeR. As a comparison of 13C and 15N NMR signals of the retinal and protonated Schiff base in HeR with those of other microbial rhodopsins, we have identified the more twisted 13-trans, 15-anti configuration. Multidimensional chemical shift correlations with MagRO NMR view resulted in the backbone assignments of over 60 amino acid residues. [1] A. Pushkarev et al. (2018) Nature, 558, 595-599.

1Pos077 (1SHP-6) 理論計算による熱安定化ムスカリン M2 受容体の選択的アンタゴニスト AF-DX 384 結合型構造
(1SHP-6) Structural insights into the subtype-selective antagonist binding to the M2 muscarinic receptor

Ryoji Suno[1], Sangbae Lee[2], Shoji Maeda[3], Satoshi Yasuda[4], Keitaro Yamashita[9], Kunio Hirata[5,6], Takeshi Murata[7], Masahiro Kinoshita[8], Masaki Yamamoto[5], Brian Kobilka[3], Nagarajan Vaidehi[2], So Iwata[8], Takuya Kobayashi[1] ([1]*Kansai Medi. Univ.*, [2]*City Hope Med Ctr.*, [3]*Stanford Univ.*, [4]*Chiba Univ.*, [5]*RIKEN,SPring-8*, [6]*JST,PRESTO*, [7]*IAE, Kyoto Univ.*, [8]*Med,Kyoto Univ.*, [9]*Univ.Tokyo,Sci*)

Human muscarinic receptor, M2 is one of the five subtypes of muscarinic receptors belonging to the family of G protein-coupled receptors. We report high resolution structures of a thermostabilized mutant M2 receptor bound to a subtype selective antagonist AF-DX 384. Comparison of the crystal structures and pharmacological properties of the M2 receptor shows that the Arg in the S110R mutant mimics the stabilizing role of the sodium cation, that is known to allosterically stabilize inactive state(s) of class A GPCRs. Molecular Dynamics simulations reveal that tightening of the ligand-residue contacts in M2 receptor compared to M3 receptor leads to subtype selectivity of AF-DX 384.

1Pos078 (1SCA-4) cDNA ディスプレイとセルソーターの利用による新規リポソームポア形成ペプチドの創製
(1SCA-4) Novel pore-forming peptides assembling in liposome membranes selected by combining cDNA display method with cell sorter system

Naoto Nemoto[1], Toshiki Miyajima[1], Takeru Yoshinobu[1], Yusuke Sekiya[2], Ryuji Kawano[2] ([1]*Grad. Sci. Eng., Saitama Univ.*, [2]*Dept. Biotech. Life Sci., Tokyo Univ. Agr. Tech*)

Previously, we have selected some liposome-binding peptides by in vitro selection using cDNA display. cDNA display method is a genotype-phenotype linking method with a cell-free translation system by fusing a cDNA with its coding polypeptide via a puromycin covalently. Owing to its stability and robustness, cell-based selections can be performed with this technology just like phage display. In this study, we performed an in vitro selection of pore-forming peptides which act functionally like antimicrobial peptides in liposome membranes, by using cDNA display with a Fluorescence Activated Cell Sorting system. Interestingly, the selected peptides form channels with ranging from 2 to 5 nm diameter in the liposome membrane by analyzing high-throughput lipid bilayer system.

1Pos079 特異的な残基置換が、水和水のダイナミクスにおよぼす影響を、解析するソフトを開発
Development of software to analyze the effects of specific residue substitution on hydration water dynamics

Ryoi Ashida, Nobuya Hasegawa, Takuya Azami, Kota Kasahara, Takuya Takahashi (*Graduate School of Life Science, Ritsumeikan University*)

The mobility of hydration water is important in determining the structure and function of proteins. Previous studies have analyzed the self-diffusion coefficient, rotational relaxation time, number of hydrogen bonds, and radial distribution function of water molecules from the calculation results based on molecular dynamics method for a specific protein. However, these analyses focused on hydration layers around the entire solute and analyzing solvation properties of a specific residue is not straightforward. To see the effects of substituted residues in more detail, we developed a software that can analyze specific regions around the residues. This software has been verified using a simple system.

1Pos080 A comparative study of membrane-bound structure of antimicrobial peptides L- and D-phenylseptin

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L-phenylseptin and D-phenylseptin are amphibian antimicrobial peptides isolated from the skin secretion of *Hypsiboas punctatus*. In the N-termini, L-Phes and D-Phes contain three consecutive Phe sequence (FFF and FfF). However, their mechanism of action and the role of the d-amino acid residue have not been elucidated yet. In this work, the interactions of both peptides with 1,2-dymyristoyl-sn-glycero-3-phosphocholine (DMPC) were investigated by means of Far-UV CD, VCD, ^{31}P NMR, and MD simulation. Both peptides form an α-helix structure in the DMPC lipid bilayers and induce similar changes in the dynamics of DMPC lipids. We found the slight difference between both membrane-bound states by MD simulation.

1Pos081* Observation of Tumor Suppressor p53 Search Dynamics using Sub-millisecond Resolved Single-molecule Fluorescence Microscopy

Dwiky Rendra Graha Subekti[1,2], Agato Murata[1], Yuji Itoh[1], Satoshi Takahashi[1], Kiyoto Kamagata[1] ([1]*IMRAM, Tohoku Univ.*, [2]*Grad. Sch. Sci., Tohoku Univ.*)

For the efficient target DNA search, DNA binding proteins utilize several microscopic motions; however, the conventional single molecule fluorescence microscopy (SMFM) cannot resolve two motions, jumping and sliding along DNA, due to the low time resolution. In this study, we developed the sub-millisecond resolved SMFM having 60-fold improved time resolution and observed the movement of a tumor suppressor p53 along DNA. The obtained single-molecule trajectories demonstrated distinct jumps of p53 along DNA in addition to the sliding along DNA. Also, the transient binding of p53 to DNA was observed. The jumping and transient binding might facilitate the target search of p53 by avoiding obstacles bound to DNA in crowded cellular conditions.

1Pos082 (1SBA-6) 大腸菌非六量体型 DNA ヘリカーゼ UvrD C 末端欠損変異体の 1 分子イメージング
(1SBA-6) Single-molecule imaging of a non-hexameric *Escherichia coli* helicase UvrD mutant lacking C-terminal residues

Hiroaki Yokota (*Biophotonics Lab., Grad. Sch. Creation New Photon. Indust.*)

Escherichia coli UvrD protein is a non-hexameric superfamily I DNA helicase which plays a crucial role in nucleotide excision repair and methyl-directed mismatch repair. We revealed by single-molecule fluorescence imaging that wildtype UvrD unwinds DNA in the form of an oligomer (dimer or trimer) (*Biophys. J.* 2013). Although it has been reported that C-terminus residues are crucial to dimerization and unwinding activity of UvrD, their role is poorly understood. Thus, in this study, single-molecule direct visualization was performed for a UvrD mutant lacking C-terminal residues. Contrary to the monomeric model proposed by biochemical, genetic and X-ray crystallography studies using the mutant, this study showed that the mutant also unwound DNA in the form of an oligomer.

1Pos083 スピンラベル ESR によるヘテロクロマチンタンパク質 HP1 の動的構造の研究：アイソフォーム特異性とリン酸化
Structural dynamics of heterochromatin protein HP1 studied by spin labeling ESR spectroscopy: Isoform specificity and phosphorylation

Toshiaki Arata[1,5], Shigeaki Nakazawa[4], Yuichi Mishima[5], Kazunobu Sato[4], Takeji Takui[4], Toru Kawakami[5], Hironobu Hojo[5], Toshimichi Fujiwara[5], Makoto Miyata[1], Isao Suetake[2,3,5] ([1]*Dept. Biol., Grad. Sch. Sci., Osaka City Univ.*, [2]*Koshien Univ.*, [3]*Twin Research Center, Osaka Univ.*, [4]*Dept. Chem., Grad. Sch. Sci., Osaka City Univ.*, [5]*IPR, Osaka Univ.*)

We reported nm-geometry and nsec-dynamics of HP1 by CW and pulse ESR. Here, the rotational dynamics of nitroxide spin label at HR in HP1α showed highly mobile on subnsec and restricted weakly with DNA in viscous glycerol, suggesting translational diffusion of HR on DNA. The CD in HP1α slowed 1.5-fold by methylated peptide (H3K9me), while that in HP1γ did not. The CSD dimer also showed isoform-specificity; 1.5-fold slower in HP1α than in HP1γ, but identical without HR. However, the chimera where HR of HP1γ was replaced with that of HP1α restored slower CSD dynamics, but not slowing of CD by DNA. Interestingly, phosphomimic mutation of NTE slowed CD or HR dynamics in HP1α but made it insensitive to DNA binding. The phosphorylated NTE may bind to HR to block DNA.

1Pos084 Protein localization in DNA cruciform junction studied by molecular simulation

Mami Saito, Shoji Takada (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

Cruciform structures are non-canonical DNA formed by inverted repeat sequences and targets for many architectural and regulatory proteins. Recently many researches reveal the role of cruciform in gene expression, replication, nucleosome structure and recombination. However, feature for cruciform binding proteins is still unclear. Using some DNA-binding proteins, we addressed feature of protein localization in cruciform performing molecular dynamics simulation with CafeMol.

1Pos085 HIV Vif–ヒト E3 ユビキチンリガーゼ複合体によるヒト抗ウイルス因子 APOBEC3G の脱アミノ化阻害の分子メカニズム
Inhibition mechanism of HIV Vif-human E3 ubiquitin ligase complex against enzymatic activity of APOBEC3G

Keisuke Kamba[1], Li Wan[1,2], Satoru Unzai[3], Ryo Morishita[4], Takashi Nagata[1,2], Masato Katahira[1,2] ([1]*Inst. of Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Energy Sci., Kyoto Univ.*, [3]*Front. Biosci., Hosei Univ.*, [4]*CellFree Sciences Co.,Ltd.*)

HIV -1 Vif completely neutralizes the anti-HIV activities of human APOBEC3G (A3G) protein. Vif forms a five-membered complex (Vif complex) by hijacking the components of human E3 ubiquitin ligase (Culin5, Elongine B, and Elongine C) and a transcription factor CBFβ. Vif reportedly binds to the N-terminal domain of A3G, leading to ubiquitination of A3G and subsequent degradation of A3G through ubiquitin-proteasome proteolysis. We investigated the effect of Vif complex on deaminase activity by A3G. We carefully inspected the interaction between A3G, Vif complex, and ssDNA using SEC, AUC, EMSA, FA, and NMR. We found that Vif complex exhibits DNA-binding activity, and forms complex with A3G and multimerizes, both of which resulted in the inhibition of deaminase activity.

1Pos086* 薬剤によるリボスイッチ“SPINACH”の構造とイオンへの影響
Influence on the structure and dynamics of Riboswitch “SPINACH” and potassium ions by DFHBI

Lisa Matsukura[1], Nobutaka Onishi[1], Masaya Furue[1], Naoyuki Miyashita[1], Takuma Shiraki[1], Yasushige Yonezawa[2] ([1]*BOST, KINDAI*, [2]*IAT, KINDAI*)

Single-molecule measurement of the intracellular molecules is useful methods to clarify the dynamics of molecules in the cell. SPINACH is one of the RNA aptamers created by mimicking GFP and exhibits a fluorescence comparable to that of GFP by binding a small molecule called DFHBI. It is useful to understand the complex RNA dynamics in the cell if the dynamics of Riboswitch and the color of the fluorescence can be controlled. However, the detail of Riboswitch dynamics has not been clear. In this study, we performed the molecular dynamics simulations of the Riboswitch “SPINACH”. SPINACH has potassium ions between and below the two G-quadruplexes with or without DFHBI. We found the dynamics of ions in the SPINACH with DFHBI are different from that w/o DFHBI.

1Pos087* シスプラチンは二本鎖 DNA を桁違いに硬くする：一分子揺らぎの定量的解析
Cisplatin causes DNA much stiffer: Quantitative evaluation viscoelasticity through the analysis of single molecule fluctuation

Toshifumi Kishimoto[1], Yuko Yoshikawa[1], Seiji Komeda[2], Kenichi Yoshikawa[1] ([1]*Grad.Sch.Life Med.Sci.,Univ.Doshisha*, [2]*Fac.Pharm.,Univ.Suzuka.Med.Sci*)

Cisplatin, a platinum-based compound, is widely used as a chemotherapeutic drug for cancer treatment. On the other hand, transplatin, the trans isomer of cisplatin, has been regarded as clinically ineffective. In this study, we quantitatively evaluated the effects of cisplatin and transplatin on the higher order structure of DNA, based on the analysis of thermal fluctuation of individual DNA molecules observed with fluorescence microscopy. It was found that cisplatin increases both the spring and damping constant on of single DNA one-order larger. In the presentation, it will be argued how such significant change of the biological function, i.e. gene expression, is caused from the specific manner of chemical interaction on the platinum isomers on genomic DNA molecules.

1Pos088 (1SEA-6) エピジェネティック修飾をもつクロマチンのモデルにおける不連続相転移
(1SEA-6) Discontinuous Phase Transition in a Chromatin Model with Epigenetic Modification

Kyosuke Adachi, Kyogo Kawaguchi (*RIKEN BDR*)

Chromatin is a complex of DNA and histone proteins. Histones are amenable to several kinds of epigenetic modifications, e.g., acetylation and methylation, which play important roles in regulating the gene expression. Interestingly, recent experiments have suggested megabase (Mb)-scale change in both the chromatin spatial structure and the histone modification profile through differentiation. However, the mechanism of such large-scale change has not been clarified. In this study, we propose a polymer model to describe a chromatin with dynamic histone modifications and find that the model can show a discontinuous phase transition. We discuss the possibility that this phase transition is the physical origin of the Mb-scale chromatin change observed in experiments.

1Pos089 トリヌクレオソームから構築するポリヌクレオソーム構造の特徴
Characterization of Poly-nucleosome Structure Constructed from Tri-nucleosome

Hiroo Kenzaki[1], Shoji Takada[2] ([1]*Info. Sys. Div., ISC, RIKEN*, [2]*Dept. Biophys., Grad. Sch. Sci., Kyoto Univ.*)

We had performed the conformational search of tri-nucleosome by coarse-grained simulation, and various basic structures were obtained deeply depend on the length of linker DNA. From these simulations, tri-nucleosome is likely to a basic unit of poly-nucleosome structure. In this study, we will construct the structure of tetra- and penta-nucleosomes by connecting the tri-nucleosome structures. Especially, we will investigate the case where the same linker length is repeated and clarify the conformational characterization at each linker length.

1Pos090 局所変形下における絡まりあった DNA 溶液の緩和過程
Relaxation process of entangled DNA solution after local deformation

Akinori Miyamoto, Yoshihiro Murayama (*Department of Applied Physics,Tokyo University of Agri. and Tech.*)

Viscoelasticity of polymer solution is important to understand chemical reactions in a cell filled with biopolymers. Especially, entangled polymer solution induces non-trivial viscoelasticity which causes anomalous diffusion, and can directly affect the reaction rate. Recently, we found that entangled DNA solution shows non-exponential relaxation even in linear response regime, which cannot be explained by only reptation theory. To elucidate the relaxation process of entangled DNA solution, we investigated the relaxation process after local deformation by using optical tweezer and fluorescence observation of DNA. We will discuss the relation between the relaxation process and concentration gradient of entangled DNA.

1Pos091 In-cell NMR 法を用いたヒト生細胞内核酸の構造およびダイナミクスの評価
Evaluation of the structure and dynamics of nucleic acids inside the living human cells by in-cell NMR spectroscopy

Yudai Yamaoki[1], Takashi Nagata[1,2], Ayaka Kiyoishi[2], Masayuki Miyake[2], Fumi Kano[3], Masayuki Murata[3,4], Masato Katahira[1,2] ([1]*Inst. Adv. Energy, Kyoto Univ.*, [2]*Grad. Sch. Energy Sci., Kyoto Univ.*, [3]*Inst. Innovative Res., Tokyo Inst. Technol.*, [4]*Grad. Sch. Arts and Sci., Univ. Tokyo*)

The properties, such as structure, stability, and dynamics of nucleic acids can be different between dilute aqueous solution and intracellular crowded conditions. In-cell NMR is a robust method for the direct measurement of those properties in the living cells. However, there are only a few reports of in-cell NMR study on nucleic acids. Here, we introduced DNAs and RNAs into living human cells using streptolysin O, which is a reversible pore forming toxin, and observed the NMR signals of the DNAs and RNAs for the first time in living human cells. Analyzing the dynamics of the imino protons involved in base-paring suggested that the structural stabilities of hairpin, i-motif, and G4 structures were different between in the living human cells and in *in vitro* conditions.

1Pos092 ハンマーヘッドリボザイムの酵素反応に関する理論的研究
Theoretical study on an enzymatic reaction of the hammerhead ribozyme

Ayaka Matsuyama, Masahiko Taguchi, Shigehiko Hayashi (*Kyoto University*)

The hammerhead ribozyme is an RNA that causes an enzymatic reaction cleaving a specific phosphodiester bond in the sequence. Currently, it is presumed that in the cleavage reaction N1 of G12 and 2'OH of G8 act as the general base and the general acid, respectively, and two Mg^{2+} the active site are involved. Although various experimental and theoretical studies have been conducted, details of the reaction mechanism have not been clarified. The purpose of this study is to analyze the reaction mechanism by *ab initio* QM/MM free energy geometry optimizations. The results suggest that upon formation of a near attacking state, the Mg^{2+} migrates near the scissile phosphate and interacts with the cleavage site to facilitate the reaction.

1Pos093* 2価ポリアミンが引き起こす遺伝子発現の促進と抑制：アミノ基間の炭素鎖長の重要性
Promotion and inhibition of gene expression caused by divalent polyamines: Marked effect of the distance between amino groups

Hiroko Tanaka[1], Chwen-Yang Shew[2], Yuko Yoshikawa[1], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Grad. Sch. Life. Med. Sci., Univ. Doshisha*, [2]*Col. Chem., City Univ. New York*)

Polyamines exist in millimolar concentrations in all living cells and are considered to play important roles on various biological functions. Here, we studied the effect of the carbon chain length (from C3 to C6) of divalent polyamines on gene expression by adapting an in vitro luciferase assay. It was found that these polyamines exhibit binary effect, promotion and inhibition, for the efficiency of gene expression depending on their concentrations. Among the polyamines, C6 diamine exhibits the lowest promotion and the largest inhibition. We may discuss such specific effect of C6 diamine in terms of geometrical effect on the interaction of polyamines with double-stranded DNA based on the conformational study with AFM and also on theoretical modeling.

1Pos094* Theoretical Studies on the Conformational Stability of RA-VII complex with 60S Ribosome

Arwansyah MS[1], Yoh Noguchi[2], Takeshi Miyakawa[2], Kazutomo Kawaguchi[1], Yukio Hitotsuyanagi[3], Satoshi Yokojima[3], Ryota Morikawa[2], Masako Takasu[2], Hidemi Nagao[1] ([1]*Division of Mathematical and Physical Sciences, Graduate School of Natural Science and Technology, Kanazawa University*, [2]*School of Life Science, Tokyo University of Pharmacy and Life Sciences*, [3]*School of Pharmacy, Tokyo University of Pharmacy and Life Sciences*)

RA-VII has been reported to have anti-tumor activity due to the inhibition of protein synthesis in the ribosome. However, the conformation of RA-VII in complex with ribosome has not been reported. In this research, we perform docking and molecular dynamics simulations to investigate the possibility of RA-VII binds at the 60S ribosome. From our simulations, the sequence of Tyr-3 of RA-VII for six models makes the hydrogen bonds with 60S ribosome. Our results suggest that all models obtained by the present simulation shows a good agreement with the experimental result since the Tyr-3 plays a major role in the inhibition of ribosome, and model 6 can be the most stable conformation because the highest value of the solvation free energy is obtained from this model.

1Pos095 (1SEA-3) オリゴペプチドのアミノ酸配列は DNA compaction と転写活性に著しい違いを引き起こす
(1SEA-3) Marked Difference in DNA Compaction and Transcription is Caused by Amino Acid Sequence of Oligopeptide

Tatsuo Akitaya[1], Hiroyuki Hiramatsu[2], Hideaki Yamaguchi[3], Koji Kubo[4], Shizuaki Murata[4], Toshio Kanbe[5], Norio Hazemoto[6], Kenichi Yoshikawa[7], Anatoly Zinchenko[4] ([1]*Asahikawa Med. Univ.*, [2]*Fac. Pharm., Meijo Univ.*, [3]*Fac. Agr. Sci., Meijo Univ.*, [4]*Grad. Sch. Env. Std., Nagoya Univ.*, [5]*Grad. Sch. Med., Nagoya Univ.*, [6]*Grad. Sch. Pharm. Sci., Nggoya City Univ.*, [7]*Fac. Bio. Med. Sci., Doshisah Univ.*)

Compaction of T4 phage DNA (166 kbp) by short oligopeptide octamers composed of two types of amino acids, four cationic lysine (K), and four polar nonionic serine (S) having different sequence order was studied by single-molecule fluorescent microscopy. Efficient DNA compaction by oligopeptide octamers depends on the geometrical match between phosphate groups of DNA and cationic amines. The mechanism of DNA compaction changes from a discrete all-or-nothing coil-globule transition induced by a less efficient (K4S4) octamer to a continuous compaction transition induced by a (KS)4 octamer with a stronger DNA-binding character. Marked difference in the morphology of the folded DNA and the transcription activity were induced corresponding to the DNA compaction mechanism.

1Pos096 Long-range Electron-Electron Interaction and Charge Transfer in Protein Complexes

David Gnandt, Thorsten Koslowski (*Institute of Physical Chemistry, University of Freiburg*)

We suggest a strategy to compute the energy landscape of electron transfer in large protein systems. For small complexes, the energy of all electronic configurations can be scanned by a numerical solution of the Poisson-Boltzmann equation. Larger systems have to be treated using a pair approximation. Effective Coulomb interactions between neighboring sites of excess electron localization become as large as 200meV. They depend in a nontrivial manner on the intersite distance. We discuss the implications of strong Coulomb interactions for the thermodynamics and kinetics of charging and decharging a 28 cytochrom *c* center containing nitrite reductase $NrfH_2A_4$ of *Desulfovibrio vulgaris* with embedding the system into a biomembrane.

1Pos097 —

1Pos098 拘束条件を用いた DFTB-MD シミュレーションの高速化とエネルギー保存の評価
Accelerate simulations and assessment of the energy conservation for the DFTB-MD simulations using the constraint method

Hiroaki Nishizawa, Yasuteru Shigeta (*CCS, Univ. of Tsukuba*)

Recently, molecular dynamics (MD) simulation based on the density-functional tight-binding (DFTB) method, which is one of the semi-empirical quantum mechanics (QM) technique, becomes applying to the biomolecular systems. In the MD simulation, the amount of the simulation sampling and the energy conservation affect the accuracy of each properties. However, the computational cost of the QM based MD simulation is very large. Furthermore, there are few assessment of the energy conservation for the DFTB-MD simulations. In this presentation, we apply to the constraint technique, namely RATTLE, to DFTB-MD simulation to accelerate simulations, and verify its energy conservation. Furthermore, we also report the matrix form of RATTLE to quickly sufficient the constraints.

1Pos099 光合成系 II の酸素発生中心の S1 状態での 12 の構造モデルの DLPNO-CCSD(T)法による計算
DLPNO-CCSD(T) calculations of twelve structural models for the S1 state of oxygen evolving complex of photosystem II

Koichi Miyagawa[1], Takashi Kawakami[2,5], Hiroshi Isobe[3], Mitsuo Shoji[4], Shusuke Yamanaka[2], Kazuhiko Nakatani[1], Mitsutaka Okumura[2], Takahito Nakajima[5], Kizashi Yamaguchi[1,5,6] ([1]*ISIR, Osaka Univ.*, [2]*Grad. Sch. of Sci., Osaka Univ.*, [3]*Grad. Sch. of Nat. Sci. and Tech., Okayama Univ.*, [4]*CCS, Univ. of Tsukuba*, [5]*R-CCS, RIKEN*, [6]*Inst. Nanosci. Design, Osaka Univ.*)

Relative stability between 12 structural models in the S1 state of the Kok-Joliot cycle for the oxygen evolving complex (OEC) of photosystem II (PSII) were investigated by hybrid DFT, double-hybrid DFT and beyond DFT method such as DLPNO-CCSD(T) method. These models include 5 hydroxide models and 1 oxo model for right-opened and left-opened case, respectively. In some models, almost same energy stability was indicated due to the dynamical electron correlation effect. This tendency indicates the possibility of the multi-intermediate state for S1 state of the Kok-Joliot cycle for the OEC of PSII. Furthermore, the implications of present results are discussed to elucidate scope and applicability of modified double hybrid DFT methods.

1Pos100* 生体溶液中のアニオンサイト間実効引力が発生する最大のサイトサイズ
Maximum size for attractive anionic-sites in a biological solution

Michika Takeda[1], Ayumi Suematsu[2], Ryo Akiyama[3] ([1]*Graduate of Science, Kyushu University*, [2]*Kyushu Sanyo University*, [3]*Institute of Science, Kyushu University*)

The anionic- sites, such as negatively charged oxygen sites, attract each other under certain electrolyte concentration. Here, we discuss the attraction mediate by cations, because it seems that the effective attraction plays an important role in the various protein associations and also cell adhesion. In the present study, the dependence of effective interaction on the size and on the charge were studied to discuss behaviors in a model biological solution. The effective interaction was calculated by the OZ-HNC theory, which is the integral equation theory of liquid. We adopted the reduced effective second virial coefficient as a measure of association. The maximum size for attractive anionic-sites will be discussed in our presentation.

1Pos101 MD シミュレーションで、モデルタンパク質の水和水のダイナミクスを明らかにする
MD simulations reveal hydration dynamics around model proteins

Takuya Takahashi[1], Takuya Azami[2], Nobuya Hasegawa[1], Ryoi Ashida[1,2], Kota Kasahara[1] ([1]*Coll. Life Sci., Ritsumeikan Univ.*, [2]*Grad. Sch. Life Sci., Ritsumeikan Univ*)

In this study, MD simulations were performed on various peptides as protein model molecules and the effects of their physicochemical properties and structure on the hydration dynamics were analyzed. In order to advance the theoretical elucidation of hydration water dynamics, we proposed a new approximation function to the time correlation function of rotational movement as well as the analysis of the translational movement and the number of hydrogen bonds. Specifically, we used polyglutamic acids with different charge state and structure, and hydrophobic polypeptides as the models of the protein's intrinsically disordered region and structural region. We also evaluated the effect of water models, solvent ions and force fields on hydration dynamics.

1Pos102 All-atom molecular dynamics simulation of the reduced protein-protein interaction with metabolites in the cytoplasm

Isseki Yu[1,2], Michael Feig[3], Yuji Sugita[2] ([1]*Maebashi Institute of Technology*, [2]*RIKEN Theoretical Molecular Science Lab.*, [3]*Michigan State University*)

Inside of a cell is highly crowded with a large number of macromolecules together metabolites. Recently, ATP, one of the most abundant metabolites in cell, was found to inhibit the aggregation of intrinsic disordered proteins (IDPs). However, its molecular level mechanism is not fully understood yet. To understand the effect of ATP and other metabolites on the protein-protein interaction from the microscopic viewpoint, we performed all-atom molecular dynamics (MD) simulations of cytoplasm by changing the metabolite or ion concentration. The reduction of protein-protein interaction with the presence of metabolites is shown, and its detailed mechanism are investigated with exhaustive analysis of metabolite-protein interactions.

1Pos103 プロテインーリガンド結合における水溶媒の役割
Role of water solvent in protein-ligand binding

Yutaka Maruyama[1], Ayori Mitsutake[2], Takefumi Yamashita[3] ([1]*RIKEN R-CCS*, [2]*Dep. Phys., Meiji Univ.*, [3]*LSBM, Univ. Tokyo*)

We investigated the effect of water solvent on the molecular recognition process of proteins. First, we performed 50 molecular dynamics simulations from a state in which the ligand (IPMP) was separated from the protein (MPU-I). Then we classified a separate state and a binding state from trajectories and calculated the binding energy using three-dimensional reference interaction site model (3D-RISM) theory. The calculated binding energy is -9.6 kcal/mol, whereas the experimental value is -8.1 kcal/mol. Of the binding energy, the contribution of water solvent is -11.3 kcal/mol. In other words, it indicates that water contribution to protein-ligand binding is large. We will show the detailed analysis in the presentation.

1Pos104 Mechanical symmetry breaking in *C. elegans* dorsal-ventral axis establishment

Masatoshi Nishikawa (*Dept. Frontier Bioscience, Hosei Univ.*)

In *Caenorhabditis elegans*, the initial event of spontaneous symmetry breaking that gives rise to embryonic polarity is the midbody remnant in the two-cell embryo being off-centered, which sets the dorsal-ventral axis. This results from the asymmetric ingression of cytokinetic furrow in first cleavage, but their underlying mechanisms remain largely unexplored. Here I demonstrate that a hydrodynamic coupling between the cell cortex and cytoplasm facilitates asymmetric furrow ingression. I identified two prerequisites for this symmetry breaking: cortical contractility to drive cytoplasmic flow, and the link between the cortex and the mitotic spindle to set long-ranged cytoplasmic flow, suggesting that cytoplasmic flow influences the cytokinetic furrow ingression.

1Pos105 マウスノード不動繊毛はメカノセンサーか?: 光ピンセットによる機械刺激後の Ca^{2+}応答
Are the immotile nodal cilia in mouse embryo mechanosensors?: Ca^{2+} signaling response after mechanical stimulation by optical tweezers

Takanobu A Katoh, Katsutoshi Mizuno, Hiroshi Hamada (*BDR, Riken*)

Nodal immotile cilia, hair-like protrusions on the surface of cells, sense a flow-dependent signal to determine the Left-Right patterning of the embryo. The Ca^{2+} channel Pkd2 is required for the sensing, however, it is still controversial how cilia sense the flow. To examine the mechanosensor hypothesis, we applied mechanical stimulation to nodal immotile cilia of a Tg mouse expresses Ca^{2+} indicator GCaMP6. Intraciliary Ca^{2+} response was observed after multiple bending (39% in $n = 33$), while the response rate was decreased under the movement restriction of cilia (23% in $n = 35$) or under the treatment of $GdCl_3$ (inhibitor of channel, 9% in $n = 34$). These results support the idea that mechanical stimulation trigger the intraciliary [Ca^{2+}] response in nodal immotile cilia.

1Pos106 ライブセルイメージングが切り開くシアノバクテリアのヘテロシスト分化空間パターン維持機構
Live cell imaging sheds light on the maintenance mechanism of pattern of cyanobacterial cell differentiation

Shun-ichi Fukushima[1], Takeharu Nagai[1], Shigeki Ehira[2] ([1]*The Institute of Scientific and Industrial Research, Osaka University*, [2]*Department of Biological Sciences, Graduate School of Science, Tokyo Metropolitan University*)

Vegetative cells possess an ability to differentiate into heterocysts which perform nitrogen-fixation in the filamentous cyanobacterium *Anabaena* sp. strain PCC 7120. After the unbranched filaments consisting of a few hundred of cells face nitrogen deprivation, all of vegetative cells don't differentiate into heterocysts, but semi-regulated spatial pattern is formed; a heterocyst differentiates in the approximate center of 20-30 vegetative cells. Our time-laps observation revealed that *pknH*, which encodes a Ser/Thr kinase, is necessary for the spatial pattern maintenance. In this presentation, we will discuss the relationship between PknH and HetN which is estimated to diffuse from heterocysts to neighbor vegetative cells and laterally inhibits the differentiation.

1Pos107 細胞性粘菌における既知のシグナルを用いない細胞分化状態の検出
Detection of cell differentiation states without known signals in *Dictyostelium*

Yusuke V. Morimoto[1,2], Takuro Kawada[1], Masahiro Ueda[2,3] ([1]*Fac. Comp. Sci. and Sys. Eng., Kyushu Inst. Tech.*, [2]*BDR, RIKEN*, [3]*Grad. Sch. Frontier Biosci., Osaka Univ.*)

Cell slime mold *Dictyostelium discoideum* has been studied well as a model organism for cell differentiation. *Dictyostelium* shows simple cell differentiation into spore and stalk cells. Previous reports suggest that cAMP works to induce spore cell differentiation. However, recently, we have shown that detection of cAMP in the multicellular phase is difficult due to the decrease in the contribution of cAMP after the multicellular phase. In this study, we tried to detect the cell differentiation states by cell dynamics and signals other than cAMP. Then we were able to distinguish the differentiation state by using cell dynamics, intracellular pH and autofluorescence.

1Pos108 (1SGA-2) 心筋細胞に備わる収縮リズム恒常性の分子機構の解明
(1SGA-2) Elucidation of molecular mechanism of contraction rhythm homeostasis in cardiac myocytes

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We found that when the cardiomyocytes are warmed using the an infrared laser, the sarcomere inside the living cardiomyocytes contracts and oscillates only while warming. We named this contractive oscillation as Hyperthermic Sarcomeric Oscillations (HSOs). We considered that the properties of HSOs are likely to be important for the heart. To understand the molecule dynamics, we analyzed the sarcomeric oscillations. As a result, in spite of the large amplitude changes of the oscillations by the calcium concentration, the frequency at each period of HSOs was kept constant. That is, HSOs are equipped with Rhythmic Homeostasis. By the multi-scale cardiomyocyte simulation model, we succeeded in reproducing the HSOs with the presence of myosin and sarcomere populations.

1Pos109 トロポニン T の E244D 変異による細いフィラメント構造変化の X 線小角散乱解析
Structural changes of cardiac thin filaments caused by E244D mutation of troponin T observed by small-angle X-ray scattering

Tatsuhito Matsuo, Satoru Fujiwara (*Inst. Quant. Life Sci., QST*)

To investigate the structural changes of thin filaments caused by a cardiomyopathy-causing mutation E244D of troponin (Tn) T, small-angle X-ray scattering measurements were carried out on thin filaments containing the human cardiac wild-type Tn (WTF) and those containing the E244D mutant of Tn (DTF) in both the -Ca^{2+} and +Ca^{2+} states. Analysis by model calculation detected quaternary structural changes in both states. In particular, tropomyosin (Tm) of the DTF exposed more myosin-binding sites on actin than the WTF in the +Ca^{2+} state. Our results suggest that myosin binding is promoted by the E244D mutation, which leads to the increased force development as reported for this mutation.

1Pos110 分子動力学計算によるミオシンの第二リン酸結合部位の検証
Validation of second phosphate binding site in myosin studied by molecular dynamics simulation

Kouei Uchida, Jun Ohnuki, Takato Sato, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

Myosin converts the chemical energy of ATP hydrolysis into mechanical work along an actin filament. Recently, a novel crystal structure of myosin in ADP-Pi state was solved, in which Pi is positioned at a site different from the ATP binding site (2nd binding site). It is also suggested that the translocation of Pi increases the actin-myosin binding affinity and initiates the powerstroke of the lever-arm region. However, it is unclear whether this 2nd binding site actually exist in aqueous solution. Here we conducted molecular dynamics simulations to validate the existence of the 2nd binding site. Then we investigate how Pi affect the actin-binding region and the lever-arm region from via the dielectric allostery which we observed upon ATP binding.

1Pos111 ミオシンのアクチンに対する結合親和性の低温における弱化を説明するための統計熱力学
Statistical thermodynamics on the weakening of binding affinity of myosin for actin at low temperatures

Tomohiko Hayashi, Masahiro Kinoshita (*Inst. Adv. Energy, Kyoto Univ.*)

It is experimentally known that the binding affinity of myosin for filamentous actin (F-actin) is counterintuitively weakened at low temperatures. Using our recently developed, accurate method for assessing the hydration properties of a polyatomic solute, we identify the physical factors driving or opposing the binding, evaluate their relative magnitudes, and analyze their temperature dependences. Molecular models are employed for water and the structures of F-actin, myosin S1, and their complex are taken into account at the atomistic level. Upon binding, the water crowding in the entire system becomes less significant, leading to a large water-entropy gain. At low temperatures, however, this effect becomes smaller, which is a principal cause of the weakening.

1Pos112 1 分子・多分子実験から迫る、心機能に適した心筋ミオシンの性質
Reverse stroke of cardiac myosin revealed by single molecule microscopy is essential for heart function

Yongtae Hwang[1], Takumi Washio[2], Hideo Higuchi[1], Motoshi Kaya[1] ([1]*Department of Physics, The University of Tokyo*, [2]*Department of Human and Engineered Environmental Studies, The University of Tokyo*)

In order to elucidate how dynamics of cardiac myosins contribute to heart function, we measured forces of cardiac myosin filaments using optical tweezers and revealed stepwise displacements of actin filaments. Compared with skeletal myosin, cardiac myosin shows more frequent backward step. We further performed the simulation to reveal which molecular properties affect on the stepping behaviors, suggesting that reverse stroke is a key feature to cause frequent backward steps and higher force. Therefore, we further investigated whether single cardiac myosin can execute the power and reverse strokes using optical tweezers. Results showed trapped beads' positions were occasionally switched between two discrete levels, revealing the execution of power and reverse strokes.

1Pos113* 負荷に依存した細菌べん毛モーター回転速度の調節機構
Load-dependent speed regulation of the bacterial flagellar motor

Tsubasa Ishida[1], Myu Yoshida[2], Tohru Minamino[3], Yoshiyuki Sowa[1,2,4] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [4]*Res. Cent. Micro-Nano Tech., Hosei Univ.*)

The bacterial flagellar motor consists of a rotor and multiple torque-generating stator units. To reveal the dynamic properties of the motor at very low load, we monitored its rotation by attaching a 60 nm gold bead to the straight flagellar hook via Cys residues. The motor showed stable rotation at ~300 Hz and multiple speed levels when stator expression level was high and low, respectively. A motor of a cell lacking stator proteins showed discrete speed increments by inducing the expression of the stator genes. These results indicate that the motor speed at very low load depends on the number of active stator units. We also investigated the effects of FliL and YcgR, and mutations of peptidoglycan-binding site of stator units on motor speeds at very low load.

1Pos114* F_1-ATPase の阻害状態解析
Analysis of the inhibited form of F_1-ATPase

Sougo Mori, Hiroshi Ueno, Hiroyuki Noji (*Noji laboratory, Department of Applied Chemistry, Graduate School of Engineering, University of Tokyo*)

F_1-ATPase (F_1) is a catalytic rotary motor of ATP synthase. In order to prevent the wasteful consumption of synthesized ATP, various modes of rotation inhibition of F_1 (ADP-Mg, IF_1, ε, ζ, and so on) exists to prevent the rotation to the ATP hydrolysis direction. However, the detailed mechanisms how F_1 falls into and recovers from these inhibitions were not known. Here, we examined the ADP-Mg inhibition of F_1 from *Bacillus* PS3 (TF_1), bovine mitochondria (bMF_1), and *F. nucleatum* (FuF_1) by the single molecule rotation assay. The time constants of the rotation and the inhibition states were similar between TF_1 and bMF_1, while those of FuF_1 were shorter than those of other F_1s. These results help us to evaluate the differences of inhibitory mechanism of F_1 among species.

1Pos115* Na^+の存在に依存するべん毛モーター固定子 PomAB における複合体の解離
Destabilization of the complex formation allows high Na^+ conduction in the PomAB flagellar stator complex

Tatsuro Nishikino, Hiroto Iwatsuki, Taira Mino, Seiji Kojima, Michio Homma (*Div. Biol. Sci. Grad. Sch. Sci., Nagoya Univ.*)

The bacterial flagellar motor is composed of the rotor and stator, and their interaction is essential to generate torque. The stator in *Vibrio* forms a hetero-hexamer complex composed of four PomA and two PomB and works as a sodium channel. A plug region (44-58) in PomB is involved in the ion flux regulation and the plug deletion (PomBΔ41-120) causes leakage of sodium ions that inhibits cell growth. To clarify this regulation, we tried to purify the plug-deleted stator, but we realized PomB was easy to dissociate from PomA. Interestingly, the PomB-D24N mutation at the ion binding site suppressed this dissociation. These results suggests that the interaction between PomA and PomB is altered by the plug deletion and this change involves ion binding to D24.

1Pos116* Biomolecular motor driven cargo transportation by microtubules as a mechanosensor

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Mechanical stress on cell has profound influences on biological processes. Microtubules, a cytoskeletal component, have been drawing attention for their contribution in cellular mechanotransduction. In this work, we studied the dynamics of motor proteins, kinesin and dynein, along microtubules under mechanical stress. Kinesin driven transportation is slowed down by mechanical stress induced deformation of microtubules, whereas dynein showed rather robust behavior. Our results corroborate that microtubules serve as a mechanosensor for their associated motor proteins. This finding will provide further opportunity to explore the role of microtubules in mechanoregulation of cellular processes.

1Pos117* ハイブリッド F_1-ATPase の 1 分子回転観察
Rotation observation of hybrid F_1-ATPases between bacterial and mammalian ones

Ryo Watanabe[1], Hiroshi Ueno[1], Toshiharu Suzuki[2], Ryohei Kobayashi[1], Hiroyuki Noji[1] ([1]*Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*CLS., Tokyo tech.*)

F_1-ATPase(F_1) is a rotary molecular motor. Previous rotation assays revealed that the rotation properties differ between bacterial F_1 and mammalian mitochondrial F_1. However, it's not clear which subunits cause the differences between strains. To answer this question, we studied "Hybrid F_1" where subunits of bovine mitochondrial F_1(bMF_1) are replaced with the corresponding subunits of thermophilic *Bacillus* PS3 F_1(TF_1). From the rotation assays of the Hybrid F_1's, it is suggested that not only the catalytic subunit β, but also γ may cause the differences between strains while α doesn't cause the differences.

1Pos118* 生体分子群ロボットによる光制御時空間貨物輸送
Photo-regulated spatiotemporal cargo transportation by biomolecular swarm robot

Mousumi Akter[1], Jakia Jannat Keya[2], Arif Md. Rashedul Kabir[2], Hiroyuki Asanuma[3], Kuzuya Akinori[4], Kazuki Sada[1,2], Akira Kakugo[1,2] ([1]*Graduate School of Chemical Science and Engineering, Hokkaido University, Sapporo 060-0810, Japan*, [2]*Faculty of Science, Hokkaido University, Sapporo 060-0810, Japan*, [3]*Department of Biomolecular Engineering, Nagoya University, Nagoya, Japan*, [4]*Department of Chemistry and Materials Engineering, Kansai University, Osaka 564-8680, Japan*)

The establishment of biomolecular robot that can selectively load and unload cargoes in a highly efficient manner, is undoubtedly an ultimate scientific challenge. For ages, scientists are trying to mimic from nature to invent such robots and utilize for the further advancements. Here, we demonstrate that a systematic construction of photoresponsive DNA modified swarm robot as cargo transporter which can load, transport and unload cargoes efficiently and intelligently using the swarming of MTs in a systematic and spatiotemporal manner. In this biomolecular robot system, cellular proteins like MTs and kinesins act as actuators, DNA as the information processors and azobenzene as sensor. This type of transport system will produce a new era in the future nanorobotics.

1Pos119* べん毛モーターの分子軸受 LP リングのクライオ電子顕微鏡による構造解析
CryoEM structural analysis of the bacterial flagellar LP ring ~ a molecular bushing of the bacterial motor with almost no friction ~

Tomoko Yamaguchi[1,2], Fumiaki Makino[1,3], Tomoko Miyata[1], Takayuki Kato[1], Keiichi Namba[1,2,4] ([1]*Grad. Sch. FBS, Univ. Osaka*, [2]*BDR, Riken*, [3]*JEOL Ltd.*, [4]*Spring-8, Riken*)

Salmonella swims by 6–8 peritrichous flagellar filaments, each rotated by a rotary motor at its base. The LP ring acts as a bushing of the motor by surrounding the drive shaft of the motor called rod. The flagellar motor rotates at around 300 revolutions per second, and the LP ring supports this high-speed rotation with very low friction. However, it remains unclear how the LP ring functions as such a low-friction bushing to support high-speed motor rotation. To understand the mechanism, we solved the structure of the LP ring at 3.6 Å resolution by electron cryomicroscopy (cryoEM) and single particle image analysis.

1Pos120 べん毛フックの自然な構造
Structure of the native supercoiled flagellar hook

Takayuki Kato[1], Tomoko Miyata[1], Fumiaki Makino[1,2], Peter Horvath[1], Keiichi Namba[1,3] ([1]*Front. Bio., Osaka univ.*, [2]*JEOL*, [3]*BDR, Riken*)

Bacteria swim in viscous liquid environments by using the flagellum. The flagellum is composed of about 30 different proteins and can be roughly divided into three parts: the basal body, the hook and the filament. The hook is a relatively short axial segment working as a universal joint connecting the basal body and the filament for smooth transmission of motor torque to the filament. The native supercoiled hook structure was solved at 3.1 Å resolution by cryoEM single particle analysis. We clarified the mechanisms for its bending flexibility and rotation stability as a universal joint.

1Pos121 細菌べん毛基部体の極低温電子顕微鏡による構造解析
Structure analysis of the bacterial flagellar basal body by electron microscopy

Tomoko Miyata[1], Takayuki Kato[1], Fumiaki Makino[1,2], Yumiko Saijo[3], Keiichi Namba[1,4] ([1]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [2]*JEOL Ltd*, [3]*Grad. Sch. Med., Kobe Univ.*, [4]*BDR&SPring8, RIKEN*)

Many bacteria swim by rotating flagella, which function as helical propellers driven by reversible rotary motors that are embedded in the cell membrane. The flagellum consists of three parts: the filament, the hook and the basal body(BB). The BB houses machinery for rotation and directional switching as well as for export of flagellar proteins and contains at least five parts according to their functions: the rod, the LP ring, the MS ring, the C ring, and the type III protein export apparatus. To obtain detailed structural information of the BB, we analyzed the structure by electron cryomicroscopy and single particle image analysis and obtained high-resolution 3D maps of the BB. We will report structural details of the S-ring, the rod and parts of the export apparatus.

1Pos122 Structure of motor evolved by combination of F-ATPase and phosphoglycerate kinase for *Mycoplasma mobile* gliding

Takuma Toyonaga[1], Takayuki Kato[2], Akihiro Kawamoto[3], Tasuku Hamaguchi[4], Keiichi Namba[2,4,5], Makoto Miyata[1,6] ([1]*Grad. Sch. Sci., Osaka City Univ.*, [2]*Grad. Sch. Front. Biosci., Osaka Univ.*, [3]*IPR, Osaka Univ.*, [4]*SPring-8, RIKEN*, [5]*BDR, RIKEN*, [6]*OCARINA, Osaka City Univ.*)

M. mobile, a fish pathogenic bacterium, glides on solid surfaces by a unique mechanism. The gliding force is believed to be generated by a motor evolved from F-ATPase, in which two hexamers similar to F_1-ATPase are paired by a frame and then assembled through eight arm-like extensions to form a sheet. In this study, we determined the motor structure by single-particle electron cryomicroscopy at 6.4 Å resolution. Each of the four arms was assigned to phosphoglycerate kinase (PGK), a glycolytic enzyme, suggesting that the motor evolved by combination of F-ATPase and PGK. The asymmetric ring of the hexamer and the central shaft penetrating each hexamer suggest a rotary mechanism analogous to that of rotary ATPases, which may be responsible for gliding force generation.

1Pos123 コンデンシン分子モーターの DNA カーテン測定と構造モデリング
DNA curtain assay and structural modeling of condensin molecular motor

Hiroki Koide, Shoji Takada, Tsuyoshi Terakawa (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

Chromatin condensation, which prevents entanglement of chromatin fibers during cytokinesis, is one of the critical events in mitosis. Recently, it was proposed that condensin proteins translocate along DNA using ATP hydrolysis energy and drive the chromatin condensation process via the molecular motor activity. However, the molecular mechanism of the condensin translocation has not been uncovered. To reveal the mechanism, in this study, we measured the step size of the condensin translocation along DNA with the fluorescent microscopic method called "DNA curtain". Also, we modeled the quaternary structure of condensin by homology modeling of each domain and inter-subunit contact prediction based on the dynamic coupling analysis and experimental data.

1Pos124 nanodisc-Vo の再構成条件検討と単粒子解析
The Investigation of reconstruction and Single Particle Analysis of nanodisc-Vo

Aya Furuta[1], Jun-ichi Kishikawa[1], Takayuki Kato[2], Atsuko Nakanishi[1], Kaoru Mitsuoka[3], Ken Yokoyama[1] ([1]*Div. Life Sci., Kyoto Sangyo Univ.*, [2]*Grad. Sch. Frontier Biosci.,Osaka Univ.*, [3]*Res. Ctr. UHVEM., Osaka Univ.*)

Bacterial V type ATPase (VoV1) is a rotary molecular motor consisting of hydrophilic ATP-driven motor V1 and membrane embedded Vo powered by proton motive force. We previously obtained structure of VoV1 by single particle analysis using cryo-EM, but the resolution of Vo was too low to discuss its side chain probably due to the intermolecular flexibility between V1 and Vo. Here, we performed structure analysis of Vo reconstructed into nanodisc to improve the resolution. We optimized the solubilization conditions (detergent, lipid and scaffold protein). EM images of nanodisc-Vo were collected using CRYOARM equipped with K2 summit detector, and we performed single particle analysis by RELION. Finally, we obtained 3D reconstruction map of Vo at near atomic resolution.

1Pos125 クシクラゲの櫛板はシンクロトロン放射光による繊毛軸糸の構造・機能解析に最適な試料である
Synchrotron radiation X-ray diffractionv reveals the highly ordered structure of axonemes in the comb plate of ctenophores

Hiroyuki Iwamoto[1], Kei Jokura[2], Kazuo Inaba[2] ([1]*SPring-8, JASRI*, [2]*Shimoda Marine Res. Ctr., Univ. Tsukuba*)

The X-ray diffraction technique has a potential to image non-crystalline biological specimens at a near-atomic resolution. To do this, both the amplitude and the phase of the diffraction pattern are required, but the phase is lost when the pattern is recorded by a detector. This “phase problem” has been the largest problem for the X-ray diffraction technique. However, this situation has changed by the recent developments of algorithms to recover the once-lost phase information and restore the structure of the sample in the real-space. This technique is called “lenseless imaging”. In this talk the recent development of “lenseless imaging” is reviewed and the possibility of its application to the imaging of non-crystalline biological specimens is discussed.

1Pos126 Unidirectional-rotation and helical-motion of a cargo on a microtubule indicate torque generation and biased sideward binding of kinesin

Mitsuhiro Sugawa, Yohei Maruyama, Masahiko Yamagishi, Junichiro Yajima (*Grad. Sch. Arts and Sciences, Univ. Tokyo*)

Various kinesin constructs exhibited helical motion around a microtubule, but its mechanism remains elusive. Here we performed a comparative study of the motility of three kinesins; KIF1A, ZEN-4 and single-headed kinesin-1. Utilizing a gold-nanorod (GNR) imaging for particle tracking and polarization measurement, we found that the kinesin-bound GNRs rotate 180 degree about the short axis in one period of the helical motion. Theoretical analyses and the Monte Carlo simulations indicates that rotation of a cargo driven by kinesin motors enhances biased sideward-binding events on the microtubule lattices.

1Pos127 ウシミトコンドリア由来 F_1-ATPase の 1 分子回転解析
Single-molecule analysis of bovine mitochondrial F_1-ATPase

Ryohei Kobayashi, Hiroshi Ueno, Hiroyuki Noji (*Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*)

F_1-ATPase (F_1), one of the best-characterized molecular motors, has been studied by both single-molecule studies and structural analysis. However, the model F_1's for single-molecule studies and structural analysis have been different. So, the correlation between crystal structures and rotational pauses has been still unclear. Here, we conducted single-molecule rotation assay of bovine mitochondrial F_1 (bMF$_1$) of which many crystal structures are available. Previously, we revealed that ATP hydrolysis reaction occurs at ~80 degrees after ATP binding pause in bMF$_1$. Here, we show some new findings of angle-dependence of elementary reactions, and inhibited states by various inhibitors, which leads to much better understanding of chemo-mechanical coupling rotation of F_1.

1Pos128 二つの頭部のつながり方がミオシン VI の歩行運動に与える影響
Effects of interhead connection on the stepping motion of myosin VI

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Myosin VI shows stepping motion along actin filament with a large and variable step size. As the neck domain of myosin VI is short, this large and variable step size is not fully explained by the lever-arm model, which calls for the consideration of the effect of Brownian motion. Using the coarse-grained model as we have used for myosin II (Nie et al., PLoS Comput. Biol. 2014), we have calculated the free energy landscape for the Brownian motion of the leading head connected with the rear head bound to the actin filament. Spudich model of interhead connection is more consistent with the large and variable step size than Sweeney model. The distribution of the direction of the leading head is also affected by the choice of the interhead connection.

1Pos129 Allosteric Regulation of V_1-ATPase by Designing Non-catalytic Interface

Takahiro Kosugi[1,2,3], Tatsuya Iida[2], Mikio Tanabe[4], Ryota Iino[1,2], Nobuyasu Koga[1,2,3] ([1]*Institute for Molecular Science*, [2]*SOKENDAI*, [3]*ExCELLS*, [4]*KEK*)

Many of the complex-forming proteins exert functions by cooperative works between their constituent proteins. One of the approaches to control over the cooperative function is designing an allosteric site. Here, we attempted to allosterically regulate rotational speed of a rotary molecular motor V_1-ATPase, by designing ATP binding site in the non-catalytic interfaces. Rotational speed of the designed V_1-ATPase is faster than the native one, depending on ATP binding affinity at the designed site. The solved crystal structures and single-molecule experiments revealed that the acceleration is caused by ATP bindings to the designed site inducing ADP release in the catalytic interfaces. This strategy will open up an avenue to control over proteins allosterically.

1Pos130 キネシン-1 人工多量体のプロセシビティと一方向運動性の評価
Evaluation of processivity and unidirectionality of artificial kinesin-1 oligomers

Kimitoshi Takeda, Akihiko Nakamura, Jun Ando, Ryota Iino (*Institute for Molecular Science*)

Kinesin-1 is a dimeric motor protein that moves along microtubule. For its highly processive unidirectional motion, coordination between two motor domains through neck-linker regions is required. To understand effect of number of motor domains on the motion of kinesin-1, here we investigated motion of artificial kinesin-1 oligomers in which the motor domain/neck-linker region fused with SpyTag was covalently connected to SpyCatcher002 oligomers from a trimer to a heptamer (Khairil Anuar, Nat Commun 2019). We first observed a trimer and a hexamer, and found that only small fraction showed unidirectional processive motions, and most of them showed processive but back-and-forth motions. Now we are analyzing the motions in detail and preparing other oligomers.

1Pos131 DNA ナノフィラメント上を移動する改変型ダイニンを用いた人工輸送システムの創生
Creating artificial transport systems by using engineered dynein that moves along DNA nanofilament

Ryota Ibusuki[1], Tatsuya Morishita[1], Akane Furuta[2], Kazuhiro Oiwa[1,2], Hiroaki Kojima[2], Ken'ya Furuta[2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICR. Res. Ins., NICT. Kobe*)

Linear biomolecular motors are remarkable machines that convert chemical energy into movement. Yet the essential mechanisms remain unclear, making it difficult to customize them for microscopic actuators. A limitation is that neither motors nor tracks can be easily re-designed. Here we describe a bottom-up approach of dynein and DNA-binding proteins to create new hybrid DNA motors that move along DNA nanostructures. The hybrid motors were able to act as a cargo transporter on immobilized DNA nanotubes that contain specific DNA sequences. Further, we developed another hybrid DNA motor that recognizes a different DNA sequence to make an orthogonal set of DNA motors, allowing to construct a molecular sorter, a concentrator, and logic gates in the nanoscale environment.

1Pos132 DNA 上を動く改変ダイニンを用いて結合性と運動速度の関係を系統的に調べる
Systematic studies on the relation between binding kinetics and speed of movement using engineered DNA-based dynein motor

Tatsuya Morishita[1], Ryota Ibusuki[1], Akane Furuta[2], Kazuhiro Oiwa[1,2], Hiroaki Kojima[2], Ken'ya Furuta[2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst NICT*)

We have developed a prototype of new molecular motor systems in which a DNA-binding domain and dynein were combined to make a new motor that moves on DNA nanotubes. However, it is unknown how to design a motor that move faster or longer along the track. Here we tested diverse types of DNA-binding domains as interfaces to the DNA track. We found that 13 out of 28 types of DNA-binding domains successfully mediated DNA nanotube movement with different speeds ranging from 1 to 100 nm/s. Importantly, these variation can be attributed to the difference in the interface to the track, allowing systematic studies on the relation between binding kinetics and speed of movement. Our study would provide important clues about the mechanisms of naturally-occurring biomolecular motors.

1Pos133 Fo 回転モーターにおける固定子ー回転子間相互作用の自由エネルギー地形
Free energy landscape for stator-rotor interaction in Fo rotary motor

Dan Parkin, Daiki Yamakoshi, Genya Nakagawa, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

The Fo rotary motor is the membrane-embedded part of ATP synthase, which is composed of a stator and a rotor. Fo couples the proton translocation across the membrane with the rotor's rotation. It is considered that the rotation is driven by biasing the rotor's Brownian rotation through the proton motive force and two distinct half channels. In our previous research, we observed the rotor's Brownian rotation is biased by an electrostatic interaction between the stator and the rotor. Here, we analyze the free energy landscape for the stator-rotor interaction by conducting umbrella sampling to clarify the source of the bias in the electrostatic interaction.

1Pos134 ミオシン頭部の自由エネルギーランドスケープのスイッチングを考慮した筋収縮の三状態モデル
Three-state model of muscle contraction with switched free energy landscapes for myosin heads

Kaima Matsuda, Masaki Sasai, Tomoki P. Terada (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

Muscle contraction is generated by the relative sliding of actin and myosin filaments. With coarse-grained molecular dynamics calculation of actomyosin system, we have previously proposed that free energy landscape experienced by myosin head switches from symmetric one to asymmetric one, corresponding to the weakly bound state and strongly bound state, respectively (Nie et al., PLoS Comput. Biol. 2014). In this study, we have reproduced two-state model of myosin heads diffusing on free energy landscape with 36 nm period (Marcucci & Yanagida, PLoS One 2012) and extended this model to three-state model by incorporating the weakly bound state. Comparison between two-state and three-state model showed that efficiency is higher for the three-state model.

1Pos135 DNA オリガミを用いたミオシンフィラメントでのミオシン II モーターの高速原子間力顕微鏡による同時観察
Simultaneous observation of individual myosin II motors in DNA origami-based thick filaments by high-speed AFM

Masashi Ohmachi[1], Hiroaki Takagi[2], Keigo Ikezaki[3], Toshio Yanagida[1,4], Mitsuhiro Iwaki[1,4] ([1]*BDR, RIKEN*, [2]*Nara Med. Univ.*, [3]*Univ. Tokyo*, [4]*Grad. Sch. Front. Biosci., Osaka Univ.*)

Muscle contraction is driven by mechanical interactions of myosin II-based thick filaments with actin filaments in muscle. Simultaneous monitoring of individual myosin motors working in the myofilaments still remains a challenge. Here, we directly observed the structural dynamics of individual human skeletal myosins in DNA origami-based thick filaments by high-speed atomic force microscopy. We detected the reversible two-step lever-arm swing and its coordinated motions among neighboring myosins. Our DNA origami-based synthetic biological system will be utilized to dissect the molecular function of single molecules in biomolecular assemblies.

1Pos136 アクトミオシン運動に対するリゾチームの阻害作用
Inhibitory effect of lysozyme on the motility of actomyosin

Masaki Okami, Kuniyuki Hatori (*Grad.Sch.Sci.Eng., Yamagata Univ.*)

We focus on the movement and organization of actomyosin in the presence of proteins that possess a non-specific interaction with actomyosin because various proteins exist in living cells. Although lysozyme (basic protein) can form actin bundles via electrostatic interactions, its effect on the properties of actomyosin remains unknown. Using an in vitro motility assay, we examined the sliding movement of actin filaments on heavy meromyosins in the presence of lysozyme. At 25 mM KCl, the movement was completely inhibited at the lysozyme concentration of 0.08 mg/ml (stall concentration). As KCl concentration was increased, the stall concentration increased. This result suggests that an electrostatic binding of lysozyme to actomyosin suppressed the motility.

1Pos137 ダイナクチンサイドアームのコンフォメーション変化
Multiple conformational changes of dynactin sidearm revealed by single molecule observation

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Dynactin is a huge protein complex, which interacts with various microtubule-binding and vesicle-binding proteins, and it regulates the motility of molecular motor dynein. Although recent cryo EM studies revealed its averaged structure, our knowledge on the structure of sidearm part in dynactin complex is still limited probably because of its flexibility. Here, we report various conformations of dynactin sidearm observed by EM and AFM. We found that the sidearm could be either bound to or released from the rod part (Arp1 rod), and the dynein-binding domain in the sidearm (coiled-coil 1) adopted either a folded or an extended form, providing clues to understand how dynactin binds to microtubules and regulates dynein.

1Pos138 The contribution of microtubule-binding ability of dynactin in dynein behaviors on microtubules

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Dynactin is a dynein modulator and is a very large multi-subunit complex. We indicated that dynactin has bifacial effects on dynein motility. These effects are derived from each domain in dynactin complex. However, the regulatory mechanism by the CC1 domain and the K-rich domain in DDB complex remains unclear, and little is known about the role of the Pro-rich domain. Contrary to the previous notion that the motility of DDB complex was independent of the presence of CapGly domain, we showed that the isoform types of p150 in DDB complexes affected their motility. We are investigating the role of the microtubule-binding ability of dynactin in dynein behaviors by single-molecule observations and live cell imaging.

1Pos139* (3SDA-6) 集団細胞遊走における機械的なシグナルを介した ERK 活性伝播
(3SDA-6) ERK activation waves mediated by intercellular mechanical signals during collective cell migration

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During collective cell migration of epithelial cells, ERK MAP kinase activation propagates as multiple travelling waves from leader cells to follower cells, which orients cell movement. However, how the ERK activation propagates remains elusive. Here we found that intercellular force transmission mediates the ERK activity propagation by using FRET imaging and optogenetic control. Leader cell movement triggers extension of follower cells, resulting in ERK activation in the follower cells. The ERK activation in turn induces cell contraction, which pulls the adjacent cells, evoking another round of extension and ERK activation. Together, we reveal the principle of intercellular communication via mechanical forces underlying long-distance transmission of directional cues.

1Pos140* スピロプラズマの螺旋交換遊泳を駆動する内部リボン構造
Internal ribbon structure driving helicity-switching swimming of *Spiroplasma*

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S. eriocheiris is a helical-shaped bacterium lacking peptidoglycan layer. It swims in high viscosity solution by reversing the cell helicity from front to back. In the present study, we analyzed the internal ribbon structure by electron microscopy to elucidate the mechanism of helical reversal. The ribbon was constructed from double helix protofilaments. The protofilament was a chain of ring-like repetitive structure composed of *Spiroplasma* specific fibril protein. Pitches of the ribbon and fibril filament corresponded to that of the cell. Based on these results, we concluded that the ribbon is the swimming machinery driving the helicity-switching swimming. Structural analysis of fibril protein by electron cryomicroscopy and single particle analysis is now under way.

1Pos141* 液-液相分離における荷電性残基の影響に関する分子動力学シミュレーション
Molecular dynamics simulations to dissect effects of charged residues on liquid-liquid phase separation

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It is well known that droplets formed by liquid-liquid phase separation of disordered proteins cause many important biological effects. However, little is known for these microscopic processes and effects, especially for the effects of the charge distribution in amino acid sequence of proteins composing the droplet. In this study, we performed coarse-grained molecular dynamics simulations for higher-order assembly of protein FUS's low complexity regions and its mutants with various charge distributions. As the result, the stability of the liquid droplet decreased as the interval between each charged amino residues increases. We discuss reproducibility of the simulated phase diagram, and effects of the distributions of charged amino residues.

1Pos142* *Spiroplasma eriocheiris* の遊泳運動にかかわる細菌のアクチン MreB の重合
Polymerization of bacterial actin MreB involved in swimming motility of *Spiroplasma eriocheiris*

Daichi Takahashi[1], Makoto Miyata[1,2] ([1]*Grad. Sch. Sci, Osaka City Univ.*, [2]*OCARINA, Osaka City Univ.*)

MreB is a bacterial actin that works as a scaffold for a cell wall synthase. Novel MreBs are found in a wall less bacterium, *Spiroplasma eriocheiris*. Although general walled bacteria have one mreB gene, it has five classes of MreBs (MreB1-5). These MreBs are involved in formation and switching of *S. eriocheiris* cell helicity, which are essential for swimming. MreB3 and MreB5 were analyzed for polymerization. Electron microscopy showed that MreB3 and MreB5 formed a non-helical juxtaposed double filament and a pair of double filaments, respectively. P_i release and bulk assays showed slow polymerization dynamics as well as ATPase activity for MreB3, which are caused by substitution of a catalytic amino acid, and fast polymerization dynamics for MreB5.

1Pos143* クラミドモナス細胞の鞭毛打頻度を用いた細胞内 ATP 濃度の推定
Estimation of intracellular ATP concentration from the flagellar beat frequency in *Chlamydomonas*

Wakako Takano[1,2], Toru Hisabori[1,2], Ken-ichi Wakabayashi[1,2] ([1]*CLS, Tokyo Tech*, [2]*LST, Tokyo Tech*)

Flagellar movement is driven by ATP-dependent force generation of dyneins. Motion of detergent-extracted flagella can be reactivated by treatment with ATP, and the flagellar beat frequency (FBF) increased with ATP in a Michaelis-Menten manner. However, under conventional experimental conditions for *Chlamydomonas reinhardtii* flagella, FBF reduces at physiological ATP concentrations (~3 mM). In this study, we improved the method to reactivate the demembranated *C. reinhardtii flagella*. Our new method increased the Vmax value of the ATP-FBF curve by ~30% compared to the conventional method. By using this curve, we estimated the intracellular ATP concentration of several strains that have defects in ATP generation pathways.

1Pos144* 細菌バイオフィムの高次秩序構造
High structural order in bacteria biofilms

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In the environment, bacteria aggregate into dense social communities that are attached to submerged surfaces; these communities are known as biofilms. Biofilms begin with the attachment of a bacterium to a surface followed by division, which generates a community encased in extracellular polymeric substances (EPS). Since the cells are both self-sticky and secrete EPS, as they divide, they adhere to each other creating amorphous random structures, devoid of long-range order. Here, we describe a unique biofilm morphology grown in microfluidic channels, where the bacteria form highly structured aggregates. We determine the relationship between adhesion force and growth pressure in biofilms to investigate their mechanical properties and characterize their long-range order.

1Pos145* 合成細菌における *Spiroplasma eriocheiris* 遊泳運動の再現
Reconstitution of *Spiroplasma eriocheiris* swimming motility in a synthetic bacterium

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A crustacean pathogenic bacterium, *Spiroplasma eriocheiris* performs unique swimming motility by switching the cell helicity. We focused on six component proteins of an internal ribbon structure responsible for swimming, fibril, specific to the *Spiroplasma* genus, and five mreBs, which are actin homologs. Then, the genes of these proteins were introduced into a "synthetic bacterium", possessing a synthesized genome with only essential genes, based on the information of *Mycoplasma* species related to *Spiroplasma*. Surprisingly, the synthetic bacterium, which did not have any motility, exhibited a helical structure similar to *Spiroplasma*, and also showed the swimming motility. We are clarifying the mechanism of swimming using this motility reconstitution system.

1Pos146 (1SHA-3) Direct observation of cell mechanics under high hydrostatic pressure

Masatoshi Morimatsu, Keiji Naruse (*Grad. Sch. of Med., Dent. and Pharma. Sci., Okayama Univ.*)

Activities of daily living such as walking and mastication pressurize tissues. However, the effect of pressurization on cells at a molecular level is poorly understood due to the lack of methods that observe cells under high pressure condition. Here we used hydrostatic pressure microscope to apply high pressure to cells in vitro and investigated cellular morphology, molecules, and gene expression levels. We observed higher pressure decreased the size of cells and gene expression levels of extracellular matrix proteins were increased after pressurizing process. Our results suggest that pressurization to cells is related to maintenance of tissue function.

1Pos147 (1SHA-2) RhoA activation induces cell cycle exit and differentiation of skin cancer cells

Oleg Dobrokhotov, Masahiro Sokabe, Hiroaki Hirata (*Nagoya Univ., Grad. Sch. Med.*)

Contact inhibition of proliferation (CIP) is crucial for tissue homeostasis, while loss of CIP is a typical hallmark of cancer. Previously we have shown that actomyosin contractility is required for CIP in HaCaT keratinocytes and pharmacological activation of the RhoA-ROCK-myosin II axis in A431 skin epidermoid carcinoma cells causes inhibition of their proliferation. In the present study, further analyses reveal that activation of RhoA in A431 cells induces cell cycle exit into G0 quiescence, which is followed by their differentiation into cytokeratin-10-positive keratinocytic cells. While cancer stemness drives tumor development, induction of cancer cell differentiation through the activation of RhoA-ROCK signaling might provide a novel strategy for cancer therapy.

1Pos148 ゼブラフィッシュ胚上皮組織に内在する力が創傷治癒を制御する
Residual Stress-mediated wound healing in zebrafish epithelia

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When epithelial sheets are injured, the wounds are rapidly repaired to recover their integrity. This process is called wound healing and is known to share similar regulatory mechanisms with several biological processes. However, it is not fully understood how forces loaded on tissues contribute to wound healing. Here, we used an epithelium in zebrafish embryo as a model, and investigated contribution of tissue forces to wound healing in vivo. Our results shows that residual stress is loaded on the epithelial tissue and that forces are utilized to heal the wound rapidly. Furthermore, we measured strength of the forces by using the femtosecond laser, and found a kPa order of tissue forces are generated and utilized in the rapid wound healing.

1Pos149 TIRF 観察によるアクチン重合・脱重合における蛍光標識の影響
Effects of Dye Labels on Actin Assembly and Disassembly

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We investigated how fluorescent dyes affect actin elongation and dissociation by TIRF microscopy using actin labeled with Rhodamine (TMR) or Oregon Green (OG) conjugated to Cys374, or Alex488 (Alexa488) or ATTO520 conjugated to a lysine. Only TMR-actin slowed down the association rate as increasing the ratio of labeled-actin. Spontaneous dissociation rates of OG- and TMR-actin were same as ADP-Pi- and TMR-actin, respectively. The dissociation rate of Alexa488-actin increased over time up to the rate of ADP-actin, suggesting that Pi dissociation is influenced, since Pi in the buffer slows the dissociation of Alexa488- ADP-actin. Dye labeling have different effects on the dissociation rate of actin filaments, likely due to their Pi release.

1Pos150 変異リアノジン受容体の分子動力学シミュレーション
Molecular dynamics simulation of mutant ryanodine receptors

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Mutations in the type1 ryanodine receptor (RYR1) gene cause severe muscle diseases, such as malignant hyperthermia (MH). We studied molecular dynamics (MD) simulation of RYR1 bearing disease-associated mutations at the N-terminal region. MD simulation of the mutant RYR1 revealed that alterations of hydrogen bonds/salt bridges between N-terminal subdomains strongly correlate with the channel function of RYR1. In particular, mutations of R402, which plays a key role in connecting the three subdomains (A, B, C) of the N-terminal region, cause clockwise rotation of B and C subdomains with respect to the A subdomain. This movement might increase the open probability of the channel, explaining the basis of MH in R402 mutants.

1Pos151 脱重合が引き起こす接着斑周囲におけるアクチン細胞骨格の向きの変化
The change of the direction of F-actin caused by the filament disassembly around focal adhesions

Kiyoshi Tohyama[1], Sawako Yamashiro[2], Naoki Watanabe[1,2] ([1]*Department of Pharmacology, Kyoto University Faculty of Medicine*, [2]*Laboratory of Single-Molecule Cell Biology, Kyoto University Graduate School of Biostudies*)

We developed a new method based on single-molecule fluorescent microscopy to visualize the direction of F-actin in living cells. We labeled the both ends of tropomyosin with different fluorescent dye as a probe to monitor the direction of bound filaments and checked the orientation change of them around focal adhesion(FA)s. We found that around the tip region of FAs, F-actin rotated inwardly. By analyzing the retrograde flow of filaments around the FA, we found that they are attracted toward the FAs and depolymerized near the frontal tip of them, where FH2 region of formins is activated by released free G-actin. These results elucidate the role of FA as the site for the change of the direction and the reorganization of F-actin.

1Pos152 クラミドモナス鞭毛の根元に局在するマイナータイプダイニンの機能解析
Functional analysis of minor-type axonemal dyneins located to the basal region of *Chlamydomonas* flagella

Tomohiro Komatsu, Yusuke Kondoh, **Toshiki Yagi** (*Dept. Life Sci., Pref. Univ. Hiroshima*)

Eukaryotic flagellar movements are based on the microtubule sliding powered by dyneins. Dynein heavy chains (DHCs) are the most important subunits in dynein molecules. *Chlamydomonas* has 15 different axonemal DHCs, three of which locate to the flagellar basal region. To understand the function of the minor-type dyneins (DHC3, DHC4, and DHC11), we had isolated the mutants missing the respective DHCs, and found that the single DHC mutants displayed WT-like motility. Here, we made double and triple mutants lacking various combination of the three DHCs. The double mutant missing DHC4 and DHC11 displayed WT-like motility, but in contrast, the other mutants swam at 50-70% speed of WT, suggesting that DHC3 performs more important roles to generate flagellar bending.

1Pos153 共焦点レーザー走査型顕微鏡で捉えた軸索タンパクのブラウン運動
Brownian motions of axonal proteins captured by a confocal laser scanning microscopy

Kazunari Mouri[1], Yasushi Okada[1,2] ([1]*RIKEN, BDR*, [2]*Univ. Tokyo, Grad. Sch. Sci., Dept. Phys.*)

Some neuronal axons extend over several meters long. Motor proteins, such as kinesin and dynein, enable to transport synaptic vesicles and other soluble proteins, but characteristics of motion have not been captured clearly. We developed a method which can extract the speed and direction of flow based on fluorescent correlation spectroscopy (FCS) with image processing algorithm. In the reconstructed image sequences captured by this FCS method, we find trajectories of single particles. Applying single particle tracking (SPT) analyses for these trajectories, we observed biased Brownian motions, where the molecules diffuse, but are gradually transported to one direction. We apply these methods to several axonal proteins, and discuss quantitative results of them.

1Pos154 生体内光架橋によるべん毛回転子タンパク質 FliG と固定子タンパク質 PomA 間相互作用の検出
Interaction between the flagellar rotor protein FliG and the stator protein PomA in cells detected by *in vivo* photo-crosslink

Seiji Kojima, Hiroyuki Terashima, Michio Homma (*Div. Biol. Sci., Grad. Sch. Sci., Nagoya University*)

Torque of the flagellar motor is generated by the rotor-stator interaction coupled with the Na+ influx through the PomA/B stator complex. Conserved charged residues in the C-terminal domain of rotor protein FliG (FliGC) electrostatically interact with those in the cytoplasmic loop of stator protein PomA, so PomA-FliG physical interaction is key to torque generation. However this short-lived interaction was quite difficult to detect, and no one could demonstrate it so far. Here we employed in vivo site-directed photo-crosslinking technique to detect rotor-stator interaction using chimeric E. coli flagellar motor. Our preliminary data showed that photoreactive amino acid analog pBPA incorporated in FliGC at positions 281 and 288 allowed FliG-PomA photo-crosslink.

1Pos155 ケラトサイトの回転するストレスファイバ車輪の内側の核と外側の細胞膜の動き
Movement of inner nucleus and outer cell membrane of a rotating stress fiber-wheel in a migrating keratocyte

Chika Okimura, Yoshiaki Iwadate (*Fac. Sci., Yamaguchi Univ.*)

If skin is broken, epidermal cells from the sides of the wound migrate into the wound site to repair it. In fish, motile cells, termed keratocytes, play this role in the wound-healing process. Each cell is composed of a frontal crescent-shaped lamellipodium and a rear spindle-shaped cell body. In the cell body, stress fibers composed of actomyosin are stereoscopically arranged so as to surround the cytoplasm just like seams of a rugby ball. Their rolling motion contributes the propulsion of keratocytes. Here, we show the movement of nucleus which is surrounded by the rotating stress fibers and the cell membrane which should transmit the traction exerted by the rotating stress fibers to the substratum.

1Pos156 バクテリアべん毛タンパク質輸送装置のゲート機構
Gating mechanism of the bacterial flagellar protein export apparatus

Miki Kinoshita[1], Keiichi Namba[1,2,3], Tohru Minamino[1] ([1]*Grad. Sch. Frontier Biosci, Osaka Univ.*, [2]*RIKEN, Spring-8*, [3]*RIKEN, BDR*)

The flagellar protein export apparatus is composed of a transmembrane export gate complex made of FlhA, FlhB, FliP, FliQ and FliR and a cytoplasmic ATPase complex consisting of FliH, FliI and FliJ and transports flagellar proteins from the cytoplasm to the central channel of the nascent flagellar structure. The cytoplasmic ATPase complex couples ATP hydrolysis by the FliI ATPase with proton-coupled protein translocation across the cytoplasmic membrane. To clarify the gating mechanism of the flagellar protein export apparatus, we have performed mutational analysis of FliI and provide evidence suggesting that FliH couples ATP hydrolysis by FliI to the entry of export substrate into the central pore of the export gate complex through an interaction between FliI and FlhB.

1Pos157 スピロヘータの遊泳におけるべん毛回転と細胞形状の関係
Relationship between the flagellar rotation and cell shape in a swimming spirochete

Toshiki Kuribayashi, Shuichi Nakamura (*Grad. Sch. Eng., Tohoku Univ.*)

Leptospira is a spirochete bacterium possessing flagella between the outer sheath and the peptidoglycan layer. The cell body exhibits right-handed helix called protoplasmic cylinder (PC). The flagellar rotation by a basal motor deforms the cell ends into spiral-shape or hook-shape and gyrates the deformed cell ends. Moreover, the counter-torque of flagella rotates PC. The complicated cell shape and cell-body rotation are responsible for propelling the cell though, it remains unknown how they are coordinated during swimming. To elucidate the leptospiral motility, we measured the cell shape, the cell-end gyrations, and PC rotation. Our results showed a correlation between the cell shape and the cell-end gyrations, and that between the gyrations and PC rotation.

1Pos158 正のべん毛本数制御因子 FlhF は MS リング構成因子 FliF の極局在を促進する
The polar localization of FliF, composing MS-ring, is promoted by FlhF in Vibrio alginolyticus

Yuna Inoue, Seiji Kojima, Keiichi Hirano, Hiroyuki Terashima, Michio Homma (*Division of Biological Science, Graduate School of Science, Nagoya University*)

Vibrio alginolyticus has a single polar flagellum at the cell pole. Its location and number are positively regulated by FlhF. However, it is unknown how FlhF contributes to the flagellar assembly. In Vibrio, FliF cannot localize at the cell pole in the strain that lacks FlhF, and ectopic expression of FliF with FlhF enables FliF to localize to the cell pole without other basal body proteins. When expressed in E. coli, FlhF alone cannot facilitate the polar localization of Vibrio FliF, although FlhF itself can localize there. When Vibrio FliF and FliG, the C-ring component, are overproduced, FlhF can promote the polar localization of FliF in E. coli cells in some extent. We propose that FlhF recruits FliF to the cell pole and facilitates MS-ring assembly with FliG.

1Pos159 超解像顕微鏡法と単粒子追跡法による標的細胞へのエクソソーム取り込み機構の解明
Uptake mechanism of exosomes by target cells as revealed by super-resolution microscopy and single-particle tracking

Koichiro M. Hirosawa[1], Taka A. Tsunoyama[2], Yasunari Yokota[3], Kenichi G. N. Suzuki[1,4] ([1]*G-CHAIN, Gifu Univ.*, [2]*OIST*, [3]*Information Science, Gifu University*, [4]*JST-CREST*)

Exosomes are small extracellular vesicles (30-100 nm in diameter), which are drawing an extensive attention as intercellular messengers of proteins, lipids, and RNAs. Recent studies suggested that exosomes play a critical role in tumor metastasis. However, uptake routes of tumor-released exosomes by target cells are totally unknown. Here, we quantitatively characterized living cell membrane structures and domains for uptake of the exosomes by simultaneously performing PALM imaging and single-particle (exosome) tracking. We found that the tumor-released exosomes are most frequently localized on the patches of lysosome-associated membrane protein 2 (LAMP2) which is a phagosome marker. We will discuss the relationship between the uptake routes and the exosome profiles.

1Pos160 ミトコンドリアの形態と活性の間に相関はあるか？
Are there any correlations between the morphology and the activity of mitochondria?

Arima Okutani (*Tokyo University of Agriculture and Technology*)

Mitochondria change their morphology in response to the intracellular state. Although mitochondrial activity is high when mitochondria show the elongated tubular structure in a cell, it remains unclear how the morphology relates with the activity. The aim of this study is to compare the activity of elongated mitochondria with that of spherical mitochondria. To equalize the circumstances around mitochondria, we isolated mitochondria from HeLa cells and measured the morphology and the activity of individual mitochondria. In some experiments, we permeabilized the plasma membrane. Mitochondrial activity was estimated by observing the membrane potential formation and changes in the matrix pH upon addition of substrate. The detailed results will be discussed.

1Pos161 酸素への電子伝達阻害時のミトコンドリアの分極機構
Mitochondrial polarization upon inhibition of electron transfer to oxygen

Hinako Tanaka[1], Emika Shida[1], Ikuroh Ohsawa[2], Yoshihiro Ohta[1] ([1]*Grad. Sch. Biotech., TUAT*, [2]*Bioregulatory function., TMGH*)

In mitochondria, electrons are transferred through electron transfer chain to oxygen molecules. These processes are utilized to save the energy required for ATP synthesis as proton gradient. CN- is considered to suppress the formation of proton gradient, since CN- binds to and inhibits cytochrome oxidase in which electrons are transferred to oxygen. In this study, we show that the formation of proton gradient continues even after addition of KCN in cells. The proton gradient formation was observed with mitochondrial membrane potential sensitive dye. When we added oligomycin, an inhibitor of FoF1-ATPase, to cells, mitochondria were further polarized even in the presence of KCN. The mechanism under the polarization on the presence of KCN will be discussed.

1Pos162 Single-molecule imaging of PI3K in eukaryotic motile cell

Satomi Matsuoka[1,2,3], Seiya Fukushima[1], Masahiro Ueda[1,2] ([1]*Graduate School of Frontier Biosciences, Osaka University*, [2]*Center for Biosystems Dynamics Research, RIKEN*, [3]*PRESTO, JST*)

An anterior-posterior polarization of eukaryotic motile cells involves a local accumulation of phosphatidylinositol 3,4,5-trisphosphate, PIP3, on the cell membrane that arises dependently on a phosphatidylinositol 3-kinase, PI3K. We investigated how membrane localization of PI3K is regulated during the polarization process. Fluorescently labeled PI3K was visualized under a total internal reflection fluorescence microscopy. Statistical analysis of the single-molecule trajectories revealed that a membrane-binding lifetime was prolonged on the membrane where Ras was activated due to the direct interaction between PI3K and the activated Ras in part. The results suggest that a self-organized activation of Ras is directly coupled to an enhanced PIP3-producing activity.

1Pos163 温度条件に依存して変化する、大腸菌走化性適応過程の定量解析
Quantitative analysis of E. coli chemotaxis adaptation process that changes depending on temperature conditions

Hiroto Tanaka, Yasuaki Kazuta, Hiroaki Kojima (*Adv ICT Res Inst, NICT*)

Cells of E. coli show chemotaxis and adaptation in response to environment. We focus on effect of temperature condition to chemical stimulated chemotaxis / adaptation in this report. By using a tethered assay in a microchannel, we achieved quantitative measurement of time traces of chemotaxis adaptation under constant temperature solution exchange. We collected amino acid-stimulated time traces under different temperatures, and statistically analyzed adaptation process focusing on correlation between chemical species and temperature. As a result, we found the temperature dependence of adaptation different depending on the species. We will discuss chemical type-dependent correlation in accordance with signaling system.

1Pos164 細胞辺縁での Aβ の凝集は細胞運動を抑制した
Aggregation of Aβ at the cell periphery suppressed cell motility

Yusaku Chikai, Ryota Yamashita, Masahiro Kuragano, Kiyotaka Tokuraku (*Grad. Sch. Eng., Muroran Inst. Tech.*)

Alzheimer's disease is caused by aggregation and accumulation of amyloid β (Aβ) in the brain, but the mechanism of cytotoxicity is not well understood. We recently succeeded in real time 3D imaging of Aβ aggregation using quantum dot nanoprobe. To understand the mechanism of neurotoxicity by Aβ, we observed aggregation of Aβ in the presence of PC12 cells using time-lapse 3D imaging. The result showed that Aβ aggregates were formed at the cell periphery. And that place where many Aβ aggregates accumulated was suppressed cell motility such as protrusion formation. When cytochalasin D was added to the culture, the aggregation was remarkably decreased, suggesting that the Aβ aggregation at the cell periphery was related to cell motility.

1Pos165 Mechanisms of negative gravitaxis in free-swimming *Chlamydomonas reinhardtii*

Azusa Kage[1], Toshihiro Omori[2] ([1]*Dept. Mech. Eng., Toyohashi U. Tech.*, [2]*Dept. Finemechanics, Tohoku U.*)

Negative gravitaxis is observed in the unicellular green alga *Chlamydomonas reinhardtii* among other protists. We have shown that negative gravitaxis in *C. reinhardtii* originates from shape asymmetry in the cell: the flagella make the cell rotate upwards in addition to its role as propulsive organelle. The shape of flagella, i.e. flagellar waveform, is important to generate torque in negative gravitaxis. Our computation revealed that it would take more than 25 seconds for a *Chlamydomonas* cell to change direction from downwards to upwards, under the assumption that its swimming waveform does not change. Actual vertical swimming trajectories showed steep rather than slow turning. These data suggest dual mechanisms in negative gravitaxis of *Chlamydomonas*.

1Pos166 孤立した MDCK 細胞における形態および運動の多様性
Diversity in morphological and motile patterns of isolated MDCK epithelial cells

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Morphology and motility of isolated epithelial cells (Madin-Darby canine kidney; MDCK cells) were quantitatively analyzed. A variety of morphologies and motilities such as spindle shape with little motion, shuttle shape and motion, and D shape with straight traveling motion, were observed even though the samples were taken from the same cell line and the experimental conditions were constant. To show these cell behaviors, a simple mathematical model was constructed where a single cell is assumed to be composed of focal adhesions (FAs) and stress fibers connecting FAs. The results suggest that the various morphologies and motilities were self-organized even though the parameter values, describing properties of cells and their interactions with environment, were fixed.

1Pos167* リン脂質スクランブラーゼ XKR による昆虫細胞膜の高粘弾性変形能
Enhanced viscoelastic deformation of insect cell membrane by phospholipid scramblase XKR

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We analyzed the physical property of cell membrane in small insects, such as the fruit fly Drosophila melanogaster and mosquito Aedes albopictus, and found that they have highly deformable cell membranes with extremely low cell-surface tension and high resistance to mechanical stress. Furthermore, biophysical and biochemical analyses revealed that phospholipid scramblase XKR, a bidirectional phospholipid transporter between the bilayer leaflets, is constitutively active in small insect cells and is a crucial regulator for viscoelastic deformation and resistance to mechanical stress of insect cell membranes. From these results, we proposed a novel mechanism for the phospholipid-scrambling mediated regulation of viscoelastic deformability in small insect cell membrane.

1Pos168* ピコ秒時間分解けい光分光法で評価した膜標的薬物の人工脂質二重膜への影響
Effect of membrane-targeted drugs on artificial lipid bilayer membranes evaluated by picosecond time-resolved fluorescence spectroscopy

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Physiological mechanism of local anesthetics is not fully understood. While they block the Na+ transport of neuron cells by interacting directly with Na+ channels, they can also affect the function of the Na+ channels by changing the physical properties of the plasma membranes. We evaluate the viscosity of lipid bilayer membranes with picosecond time-resolved fluorescence spectroscopy, by measuring the photoisomerization rate constant of trans-stilbene embedded in the membranes. In this work, we estimate the viscosity of artificial lipid bilayers with the presence of three membrane-targeted drugs, lidocaine, lidocaine hydrochloride, and capsaicin. The results indicate that the membrane viscosity increases when capsaicin is added to the membrane.

1Pos169* ボトムアップ配列設計による α-ヘリックスペプチドナノポアの構築
De novo design for pore-forming peptides with α-helical structure

Masataka Usami, Keisuke Shimizu, Yusuke Sekiya, Ryuji Kawano (*Tokyo University of Agriculture and Technology*)

We report the design strategy for α-helical peptides, which forms nanopore in a bilayer lipid membrane (BLM). We have recently attempted to construct a programmable nanopore by the bottom-up design of pore-forming peptide. In this study, we designed amphipathic α-helix peptides based on two different helix packing motif: GxxxG and GxxxxxxG. Electrostatic interaction and π-π stacking are introduced to achieve stronger interaction between the monomers. The pore-formation of the designed peptide was measured by the channel current measurement with stable BLM by droplet contact method. As results, the designed peptides form nanopore in BLM with a various number of monomers, resulting in the various size of pore-formation.

1Pos170* 脂質膜中にナノポアを構築する β シートペプチドの De novo 配列設計
De novo design of nanopore-forming β-sheet peptide in bilayer lipid membrane

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This paper describes the de novo design of β-sheet peptides to construct a nanopore in bilayer lipid membrane (BLM). Recently, de novo design method has been developed and allowed to control structure of artificial proteins. We have focused on pore-forming proteins/peptides in BLM because structure-controllable nanopore will apply to use nanopore sensing or to construct synthetic transporter. In the previous study, pore-forming α-helix peptides have been designed, but the pore stability was not enough to use for functional nanopore. Here, we designed β-sheet peptides to construct more stable nanopore. The nanopore stability was evaluated by channel current measurement. As a result, designed nanopore was as stable as those of membrane proteins by condition optimization.

1Pos171* Investigation of local anesthetic and membrane interactions using model cell membranes

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Local anesthetics(LAs) penetrate the nerve cell membrane and bind to the sodium channel to prevent neurotransmission. LAs are closely related to the cell membrane. LAs dissolve in the membrane to change thermal stability, fluidity, permeability, and so on. Tetracaine and lidocaine were selected to analyze the effects of LAs on the membranes. The relation between the LAs and the membrane was confirmed by SEM and IC50 assay with CCD-1064sk. We also analyzed the effect of LAs at the membrane level by Polydiacetylene (PDA) colorimetric assay and electrophysiological assay of planar lipid membrane. This series of experimental results conclude that the carbon tail of tetracaine affects membrane affinity, which changes the physical properties of the membrane.

1Pos172 環状ジペプチドの安定構造における系統的な傾向と特徴
Systematic trends and features in the stable structure of cyclic dipeptides

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Cyclic dipeptides (CDPs) play essential roles in biological activity to organism ranging from bacteria to humans. In particular, CDPs have significant potential for therapeutic applications. Therefore, we aimed at exploring the physical properties of each type of CDPs. In order to find systematic characteristics derived from 210 neutral CDPs, we first searched for stable structures by using density functional theory (DFT) calculation. A correlation was found between the dipole moment values and the shapes of the side chain of CDP. Besides, by calculating the solvation free energy change, ΔGsolv, of CDPs, the stabilization energy of CDP due to the difference in the solvents was evaluated, which reflect the hydrophilicity of CDPs.

1Pos173 Ca2+-dependent high-conductance channel activity of F-ATP synthase matches the permeability transition pore

Andrea Urbani[1,2,5,6], Valentina Giorgio[2], Andrea Carrer[2], Cinzia Franchin[2], Giorgio Arrigoni[2], Chimari Jiko[3], Kazuhiro Abe[7], Janna F.M. Bogers[4], Shintaro Maeda[6], Kyoko Shinzawa[5], **Christoph Gerle**[1], Ildiko Szabo[2], Paolo Bernardi[2] ([1]*IPR*, [2]*Univ. Padova*, [3]*Kyoto Univ.*, [4]*TU Delft*, [5]*Univ. Hyogo*, [6]*Scripps*, [7]*Nagoya Univ.*)

The proposal that the mitochondrial megachannel (MMC)/permeability transition pore (PTP) forms from F-ATP synthase remains controversial. By combining highly purified, fully active bovine F-ATP synthase with planar lipid bilayer electrophysiology we demonstrate that F-ATP synthase elicits Ca2+-dependent currents matching those of the MMC/PTP. Ca2+ alone was able to induce fully reversible currents. Channel activity was insensitive to inhibitors of porin and of the adenine nucleotide translocase. These findings confirm the mitochondrial F-ATP synthase to be the molecular identity of the MMC/PTP.

1Pos174 P-SPICA: A coarse-grained force field for biological systems with a polar coarse-grained water model

Yusuke Miyazaki, Susumu Okazaki, Wataru Shinoda (*Grad. Sch. Eng., Nagoya Univ.*)

A coarse-grained (CG) force field with a polar CG water model, named P-SPICA, has been developed. The water model in the CG force field reproduces several thermodynamic properties such as bulk density, surface tension, and electric permittivity. As for lipid bilayers in the CG model, they exhibit accurate molecular area, surface tension, line tension, and distribution functions comparable to those obtained by atomistic molecular dynamics (MD) simulation. Using the CG force field based on the polar water model, we can treat the electrostatic screening effect in CG-MD simulation, and appropriately simulate morphological change of anionic lipid aggregates and membrane pore formation involving antimicrobial molecules.

1Pos175 光照射によるリポソーム内への配列選択的 DNA 輸送
Sequence selective DNA transport into liposome by photoirradiation

Shigetaka Nakamura, Nobuharu Uehara, Takashi Hasegawa, Kenzo Fujimoto (*JAIST*)

Liposomes have been used as carriers of nucleic acid drugs. In this study, we demonstrated the transportation of DNA into liposomes using ultrafast DNA photo-cross-linking for delivery of nucleic acid drugs. The DNA photo-cross-linking induced cohesion of DNA on the surface of liposome, leading to its transport into the liposome. DNA transportation into the liposomes occurred in a DNA sequence-specific manner through hybridization chain reaction. Moreover, the ratio of transported DNA was dependent on the density of negative charge identical to the phosphate group of the DNA backbone.

1Pos176 生細胞膜上における相分離の誘発と可逆的制御
Induction and reversible control of phase separation on living cell membranes

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Cells have highly organized systems to condensate biomolecules to a specific compartment for effectively exerting cellular functions. However, there was no example to report a technique to artificially induce phase separation for regulating cellular functions. In this study, we used a metal complex lipid and a metal ion to create a phase separation induction system by microdomain formation on living cell membranes. Interestingly, the microdomain formation could be reversibly controlled. The size was apparently 1-3 um in diameter, which allows us to easily observe phase separation and the border region between Ld and Lo phases on living cells. Our technology is expected to become a platform to study dynamic ordering phenomena controlling cellular functions.

1Pos177 気液界面における人工肺サーファクタント膜への微粒子の影響
Effect of sub-micron particles on a model lung surfactant monolayer at the air-water interface

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Exposure to nano- and sub-micron particles can change the biophysical behavior of the lung surfactant film. Lung surfactant is a complex mixture of lipids and proteins at the air-water interface of the alveolus and responsible for reducing the surface tension. The reduction makes the process effortless during the process of inhalation. However, the mechanisms by which inhaled nano- and sub-micron particles influences the lung surfactant film are still unknown. Here we investigate the biophysical behavior of a model surfactant monolayer containing phosphatidylcholine (PC) and phosphatidylglycerol (PG) after exposure to sub-micron particles, using isotherms, surface elastic modulus and fluorescence microscopy. The details of the results will be discussed.

1Pos178 On the condensing effects of 7β-hydroxycholesterol, 25-hydroxycholesterol, and cholesterol on DPPC bilayers

Hiroshi Takahashi, Tatsuya Hoshino (*Biophys. Lab., Grad. Sch. Sci.&Tech., Gunma Univ.*)

Some oxysterols are useful precursors of hormones, but some oxysterols are harmful, causing arteriosclerosis, neurological disease, etc. One of the functions of cholesterol (Chol) is to adjust the physical properties of lipid bilayers of biomembranes appropriately. The harmful oxysterols may not work the role. We have been investigating the effects of Chol, 7β-hydroxycholesterol (7βOH), and 25-hydroxycholesterol (25OH) on DPPC bilayers. In this meeting, we will mainly focus on the condensing effect on DPPC bilayers in a fluid-like phase. The analysis of volumetric data tentatively suggested that the strength of the condensing effect is in the order of 25 OH > Chol > 7βOH at 50°C. We will also discuss the structural effects of 7βOH and 25OH on DPPC bilayers.

1Pos179 蛍光寿命測定を用いた脂質二重膜におけるセラミド-1-リン酸の動的挙動解析
Dynamic behavior of ceramide-1-phosphate in lipid bilayers examined by fluorescence lifetime measurement

Tomokazu Yasuda[1,2], J. Peter Slotte[3] ([1]*Grad. Sch. Sci., Osaka Univ.*, [2]*Research Foundation Itsuu Laboratory*, [3]*Åbo Akademi Univ.*)

Recently, it was clarified that ceramide-1-phosphate (C1P) may function by the direct interaction through the activation of cPLA2 involved in inflammation. However, the dynamic behavior of C1P in biomembranes has not been well understood. Thus, in order to elucidate how the hydrophlic structures and properties of C1P can influence lipid interaction and lateral segregation, we chemically synthesized C1P analogs and scrutinized the properties of segregated C1P domain by time-resolved lifetime. The results suggested the hydrogen bond capability and the size and charge in phosphate group largely affect C1P/colipid interactions as well as for the formation of laterally segregated domains.

1Pos180 Molecular dynamics simulation of the mechanical properties of lipid membranes in the presence of proteins

Diego Ugarte, Shoji Takada (*Takada Lab., Dept. Biophysics, Div. Biology, Grad. Sch. of Sci., Kyoto Univ.*)

Biological membranes mechanical properties and shape depend not only in the lipid composition but also in the molecules interacting with it. Specifically, some proteins can induce different shapes on membranes by inserting into them or attaching to their surface. These proteins are important because they can facilitate different biological processes like membrane pore formation, membrane fusion, etc. In this work, we use an implicit solvent coarse-grained lipid model and the AICG2+ coarse-grained protein model to perform molecular dynamics simulations of lipid-protein systems and measure the mechanical properties of membranes in the presence of proteins.

1Pos181 EGFR 膜近傍領域のリン酸化が EGFR TM-JM 二量体構造に及ぼす影響
The impact of phosphorylation in the EGFR JM region on the dimer structure of EGFR TM-JM region

Daisuke Matsuoka[1], Yuji Sugita[1,2,3] ([1]*RIKEN Theoretical Molecular Science*, [2]*RIKEN R-CCS*, [3]*RIKEN BDR*)

EGFR is a member of RTK and involved in cell proliferation. EGFR transmembrane (TM) and juxtamembrane (JM) regions are critical for the regulation of EGFR activation states. It is reported that phosphorylation of T654 residue in the JM region destabilizes the JM dimer, and the destabilization may be linked to EGFR desensitization. However, its mechanism in the molecular level is still unknown. To get clues to elucidate the mechanism, we ran multiple MD simulations for various systems in which lipid composition or phosphorylation state were different. Then we found POPS molecules affected the TM dimer structure when T654 was phosphorylated. In the presentation, we will show interaction modes between lipid and the peptide, and discuss their effects on the dimer structure.

1Pos182 The dipole potential probed by hydrophobic ions using the contact bubble bilayer method

Yuka Matsuki[1], Masayuki Iwamoto[2], Mariko Yamatake[2], Shigetoshi Oiki[3] ([1]*Dept. Anesth & Reanmat., Univ. Fukui Facult. Med. Sci.*, [2]*Dept. Mol. Neurosci., Univ. Fukui Facult. Med. Sci.*, [3]*Biomed. Imaging Res. Center, Univ. Fukui*)

Membrane potentials consist of three kinds of potentials (transmembrane, surface, and dipole potentials). The dipole potential is generated from dipoles of phospholipid molecules. Physiological implication of the dipole potential is its effect on the voltage sensitivity of the voltage-gated channels. Here we evaluated the dipole potential probed by hydrophobic ions in the contact bubble bilayer (CBB) method. Upon a step voltage change, these molecules transfer across the hydrophobic core of the membrane towards the other side of the interface, yielding transient currents. From the voltage and concentration dependence of the transient current, the dipole potential profile inside the membrane was evaluated through an application of a simple kinetic model.

1Pos183 膜活性な両親媒性ランダムコポリマーによる脂質ナノディスクの自発形成
Spontaneous formation of lipid nanodisc by membrane active amphiphilic random copolymer

Kazuma Yasuhara, Mitsuyoshi Yuma, Jinyu Hao, Jin Arakida, Rapenne Gwenael, Jun-ichi Kikuchi (*Div. Mater. Sci., Nara Inst. Sci. Tech.*)

Among various mimetics of the native membrane, lipid nanodisc is a promising material for the analysis of membrane proteins and membrane-interacting peptides. In this study, we have developed membrane-active amphiphilic polymers that form lipid nanodiscs through the fragmentation of a membrane. We have designed and synthesized amphiphilic polymethacrylate derivatives by introducing hydrophobic and hydrophilic side chains. The activity of the membrane fragmentation depended on the amphiphilic balance and molecular weight of the polymer. The polymer was found to form nanodiscs with a diameter of several tens-of-nanometers. Additionally, we applied the obtained nanodiscs to examining the interaction of amyloidogenic peptides by spectroscopies.

1Pos184 動的および静的光散乱法によるリン脂質ベシクルの構造評価 2
Structural evaluation of phospholipid vesicles by dynamic and static light scattering techniques 2

Nobutake Tamai[1], Takeshi Nobuoka[2], Ryo Takechi[2], Masaki Goto[1], Hitoshi Matsuki[1] ([1]*Grad. Sch. Tech. Indus. Soc. Sci., Tokushima Univ.*, [2]*Dept. Biol. Tech. Sci., Col. Eng., Tokushima Univ.*)

Structural studies on phospholipid bilayer membranes have been extensively performed by employing various scattering techniques (e.g., small- and wide-angle X-ray scattering) to establish their microscopic structure (e.g., bilayer thickness and molecular packing states). By contrast, there are comparatively few studies on structural features of phospholipid vesicles at mesoscopic scales (e.g., the shape and size distribution of vesicle particles) though those structural properties are significant when they are utilized as nano-materials. In this study, we attempted to evaluate more detailed structural features of phospholipid vesicle particles on this scale, especially lamellarity, by the combination of general dynamic and static light scattering techniques.

1Pos185 脂質膜外葉のみに形成したラフト様秩序領域の物性とそれが内葉に及ぼす影響
Physicochemical properties of raft-like ordered domains formed in outer leaflet and its influence on the inner leaflet

Takuya Koga, Masanao Kinoshita, Nobuaki Matsumori (*Grad. Sch. Sci., Kyushu Univ.*)

Lipid rafts are ordered membranes domains with sphingomyelin (SM) and cholesterol (chol). Although SMs abundantly reside in the outer leaflet of cell membranes, properties of the raft membrane have been examined with symmetric model membranes consisting of ternary mixtures such as SM, chol and dioleoylphosphatidylcholine (DOPC). Here, we formed raft-like ordered (Lo) domains selectively in the outer leaflet by dope of SM to DOPC/chol fluid membrane and found that the Lo domains show a higher fluidity than that formed in symmetric membranes due to the influence of the inner leaflet. Moreover, we disclosed that the phase separation in the outer leaflet induces the phase separation in the inner leaflet. These results provide direct evidence for the interlayer interaction.

1Pos186 三種のリン脂質/コレステロール・リポソームにおけるクロルゾキサゾン捕捉量とコレステロール濃度との相関
Correlation between the amount of trap of chlorzoxazone by various phospholipid/cholesterol liposomes and their cholesterol concentrations

Shosei Kano, Hiroshi Takahashi (*Biophys. Lab., Grad. Sch. Sci.&Tech., Gunma Univ.*)

Our previous study (Biochemistry, 55 (2016) 3888) have suggested that cholesterol (Chol) in biomembranes plays some role in drug metabolism by cytochrome P450 (CYP) in endoplasmic reticulum (ER). By using chlorzoxazone (CZX), CYP-substrate dug, we investigated a correlation between the amount of trap of CZX by various phospholipid/Chol liposomes and their Chol concentrations. As a result, it was found in POPC and POPE liposomes that the trapped amount became maximum at the Chol concentration corresponding to that in the ER membrane. However, a different trend was observed for PG liposomes. In addition, the effect of Chol on the interaction between PG membranes and CZX was examined by X-ray scattering.

1Pos187 ジミリストイルホスファチジルコリン２重膜に形成された脂質様錯体ドメインの物理化学的特性
Physico-chemical properties of lipophilic complex-rich domains formed in dimyistoylphosphatidylcholine (DMPC) bilayers

Hikaru Watanabe, Yoshinao Kinoshita, Ryo Ohtani, Nobuaki Matsumori (*Grad. Sch. Sci., Kyushu Univ.*)

Dabco-C16 [Mn(N)(CN)4(dabco-C16)] (LC16) is a lipophilic complex with two hydrophobic tails and one hydrophilic headgroup. It was reported that the addition of divalent metal ion Mn2+ to the LC16/DMPC bilayers causes the LC16-rich/LC16-poor phase segregation. However, physico-chemical properties of the LC-rich phase remain unknown. Fluorescent observation showed Bodipy-PC; a marker for the disordered phase, was excluded from LC16-rich domains. Calorimetry revealed that the LC-rich phase showed higher melting transition temperature than the LC-poor phase. Thus, LC16 likely forms ordered domains, consisting of DMPC/LC16 in the molar ratio of 1:0.45. In the session, we will compare the influence of Mn2+ and Ni2+ on the physico-chemical properties of LC16-rich domains.

1Pos188 低流量電子線散乱法を用いた局所的な脂質充填構造の解析
Low-flux election-diffraction discloses the local structure of lipid membrane

Shimpei Yamaguchi, Masanao Kinoshita, Nobuaki Matsumori (*Grad. Sch. Sci., Kyushu Univ.*)

Ordered membrane domains, called lipid rafts, attract multi-disciplinal interests due to their potential involvement in important biological events. So far, physico-chemical properties of lipid rafts have been investigated by using ordered/disordered phase separation occurring in artificial membranes. However, structural information on the ordered membrane domains, whose size is less than 10 μm, remains to be known because x-ray, whose size is approx. 100 μm, hardly addresses the local structures occurring in lipid membranes. To overcome this difficulty, we applied electron diffraction with higher convergence to the lipid membranes. Consequently, we selectively obtained structural information on the ordered and disordered domains formed in binary lipid monolayers.

1Pos189* 全反射赤外分光法によるムスカリン性アセチルコリン M2 受容体のリガンド認識、活性化機構研究
Ligand recognition and activation mechanism in muscarinic acetylcholine receptor M2 (M2R) study by ATR-FTIR spectroscopy

Kohei Suzuki[1], Kodai Katayama[1], Ryoji Suno[2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Kansai Medical University*)

Muscarinic acetylcholine receptor 2 (M2) is a prototypical GPCRs that are widely expressed throughout the CNS and mediates most of the actions of the neurotransmitter acetylcholine (ACh). Recent crystal structures of the M2 have provided structural insight into two conformations between inactive and active states. However, the dynamic properties during activation remain poorly understood. Here, we examine the molecular mechanism behind the ligand-induced conformational changes in M2 for both extracellular and cytoplasmic side using ATR-FTIR spectroscopy. The first agonist binding-induced ATR-FTIR difference spectra clearly showed some vibrational band change, which will be originated from structural elements of ligand binding pocket and cytoplasmic surface.

1Pos190 コレラ菌タウリン走性受容体 Mlp37 の制御ネットワーク
The regulatory network controls expression of the taurine chemoreceptor Mlp37 in *Vibrio cholerae*

So-ichiro Nishiyama[1], Hirotaka Tajima[2,3], Shiori Onogi[2], Hiroshi Urakami[1], Ikuro Kawagishi[2,3] ([1]*Fac. App. Life Sci., Niigata Univ. Pharm. App. Life Sci.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

Vibrio cholerae, the etiological agent of cholera, is attracted to taurine (2-aminoethanesulfonate), a component of bile. We have recently found that taurine chemotaxis is much enhanced when cultured at 37°C rather than 30°C. Temperature also affects expressions of virulence genes through a regulatory network, named ToxR regulon. We found that the lack of the master transcriptional regulator ToxR allows taurine taxis even at 30°C, suggesting the involvement of the ToxR network in the regulation of *mlp37*. Assays of chemoreceptor function and promoter activity are in progress to elucidate the elaborate regulatory network for temperature-dependent modulation of taurine response of *Vibrio cholerae*.

1Pos191 大腸菌グローバル転写因子 PdhR による走化性受容体遺伝子発現調節
Expression of the major chemoreceptor genes is regulated by a global transcription factor PdhR in *Escherichia coli*

Ayano Inoue[1], Nana Ito[1], Yumeno Kawasaki[1], Eri Shiokawa[1], Hirotaka Tajima[2,3], Ikuro Kawagishi[1,2,3] ([1]*Grad. Sch.Sci.&Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

In *Escherichia coli* , the genes encoding chemoreceptors, including Tsr for serine taxis and Tar for aspartate and maltose taxis, belong to the flagellar regulon, and their transcription is under the control of the master regulator FlhDC and the sigma factor FliA (σ^{28}). However, the expression of the chemoreceptor genes might be differentially fine-tuned considering that their relative importance may be different in different environments. In this study, gel shift assays revealed that a global regulator PdhR, which regulates energy metabolism and cell division factors, bind the promoters of *tsr* and *tar*. Fluorescence measurements of strains expressing Tsr and Tar fused to different fluorescent proteins revealed that these genes are positively controlled by PdhR.

1Pos192 大腸菌細胞側面膜領域における走化性受容体クラスター形成
Chemoreceptor clustering of *Escherichia coli* in lateral regions of the cytoplasmic membrane

Nana Ito[1], Masatoshi Nishikawa[2], Yoshiyuki Sowa[1,2,3], Ikuro Kawagishi[1,2,3] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

Chemoreceptors of *Escherichia coli* form a cluster at a cell pole with a trimer of homodimers as a building unit. *In vivo* imaging has revealed that chemoreceptors first inserted into lateral regions of the cytoplasmic membrane and then diffuses toward the cell pole. On the other hand, *in vivo* cross-linking experiments have suggested that homodimers of the aspartate chemoreceptor Tar and the serine chemoreceptor Tsr form mixed trimers of dimers. Here we carried out single-molecule imaging of Tar and Tsr that are fused to different fluorescent proteins. We found that Tar and Tsr colocalize even in lateral membrane regions. Further analyses of fluorescent foci suggest that they form trimers of dimers or larger clusters.

1Pos193 細菌の可溶性走化性受容体と細胞膜貫通型走化性トランスデューサー相互作用解析
Probing interaction between a soluble receptor and a transmembrane transducer in bacterial chemotaxis

Hisashi Kubota[1], Kana Murakami[2], Hirotaka Tajima[2,3], Ikuro Kawagishi[1,2,3] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Res. Cen. Micro-Nano Tech., Hosei Univ.*)

Most bacterial methyl-accepting chemotaxis proteins (MCPs) are transmembrane proteins that regulate the cytoplasmic histidine kinase CheA in response to stimuli. We have found that in *Vibrio cholerae*, the attractant L-serine either binds directly to MCPs (Mlp24 and Mlp37) or requires a substrate-binding protein (SatA) of ABC transporter in addition to MCP (Mlp3). The latter suggests that SatA serves as a primary receptor for serine and Mlp3 as a transducer. Here we tried to test this hypothesis using photo-crosslinking technique. We first detected crosslinked products of the soluble receptor (MBP) and the transmembrane transducer (Tar, an MCP) for maltose taxis of *Escherichia coli*. We then carried out similar experiments with SatA and Mlp3 expressed in *E. coli* cells.

1Pos194* 脳脊髄液流動を想定した非平衡空間におけるアミロイドβ凝集
Amyloid β Aggregation in Non-equilibrium Space Mimicking Cerebrospinal Fluidic Flow

Akane Iida[1], Kei Unoura[2], Hideki Nabika[2] ([1]*Graduate School of Sci. and Eng., Yamagata Univ.*, [2]*Faculty of Sci., Yamagata Univ.*)

The underlying mechanism of Alzheimer's disease (AD) has been proposed that amyloid β (Aβ) peptides aggregate and accumulates on the surface of the brain cells under interstitial fluid (ISF) flow. Although ISF keeps the system to nonequilibrium open system, most experiments were done with closed condition such as the Petri dish, which makes impossible to understand its molecular mechanism of AD. Thus, we focused on the role of nonequilibrium ISF on the cause of AD. Our study confirmed that the nonequilibrium flow promoted the Aβ plaque formation through the detergent-effect of Aβ, which induced macroscopic collapse of the cell membrane. A new mechanism of Aβ plaque formation and consequent nerve cell collapse was proposed by considering the role of nonequilibrium ISF.

1Pos195* 光ピンセットを用いた AMPA 型グルタミン酸受容体分子操作における神経電気活動変化
Neuronal electrical activity induced by optical trapping of AMPA-type glutamate receptors on neurons

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AMPA-type glutamate receptor (AMPAR) is one of neurotransmitter receptors at excitatory synapses. For realizing the artificial control of synaptic transmission, we have applied optical trapping of quantum-dot (QD) conjugated AMPARs on neuronal cells. Here, we evaluated the relationship between optical trapping dynamics of AMPARs and the electrical activity in neurons. The whole cell patch clamp recordings were combined with fluorescence microscope for optical trapping. When a trapping laser was focused into QD-AMPARs on neuron, fluorescence intensity gradually increased at the focal spot, suggesting that QD-AMPARs were optically trapped. The intercellular electrical activity increased during laser irradiation, which implies that it is activated due to optical trapping.

1Pos196 (1SEP-2) Single-cell trajectory analysis of human iPS cell-derived neurons carrying a rare RELN deletion

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To reach the correct destination, newborn neurons must migrate under strictly controlled conditions. However, the migration mechanisms in human neurons remain poorly understood, because of ethical concerns against the use of living human brain. To address this, we performed live-imaging analysis using human iPS cells-derived neurons. Single-cell trajectory analysis revealed that human neurons possessed directional migration even in vitro and their direction is highly correlated with cell shape orientation. On the other hand, neurons with a rare RELN deletion demonstrated a wandering type of migration and a failure of correlation between migration direction and cell shape. Our findings provide molecular and physical bases of migration mechanisms in human neurons.

1Pos197 ミミズの短期記憶メカニズム
Molecular mechanism of short-term memory formation in earthworms

Yoshiichiro Kitamura, Akira Sakane, Hikaru Tsumita (*Department of Mathematical Sciences and Physics, College of Science and Engineering, Kanto Gakuin University*)

Molecular mechanism of short-term memory in the earthworm Eisenia fetida is investigated. We previously reported that habituation by repeated tactile stimulus to the body wall in the earthworm is induced assumedly due to via serotonin (5-HT)-nitric oxide (NO)-cGMP signaling. In this study, we investigated effect of NO-cGMP signaling on establishment of short-term memory by classical conditioning of the earthworm by weak vibration and light. Administration of NO synthase inhibitor L-NAME before conditioning impaired memory formation. From these results, it is revealed that NO signaling related to short-term memory formation by classical conditioning in the earthworm.

1Pos198* 時間依存性の環境変化認識のモデリング：光結合する矩形波発振子のネットワークがもたらす幾何学的フラストレーション
Perception of time-dependent environmental change: A toy model with photo-coupled electronic oscillators composing frustrated network

Hiroshi Ueno, Masatomo Matsushima (*Dep. Med. Info., Grad. Sch. Life Med. Sci., Doshisha Univ.*)

We adapted a very simple experimental model on the coupling of excitable/oscillatory element, by using opto-electronic circuit known as multi-vibrator composed of resistor, capacitor, Op-Amp, photo coupler. A simple network of three oscillators composing a triangle connection performs the time-dependent processing of information; each neighbouring pair exhibit anti-phase synchronisation; i.e., frustrated network. In this system, multi-stability is embedded, where each mode is generated from specific time-sequential input. Certain history of input evokes corresponding mode of entrainment in these simple oscillators. Such specificity of time-dependent mode selection will be discussed in relation to the potentiality of perception with coupled oscillators.

1Pos199 大脳皮質神経細胞の単一配置による神経回路の構築
Construction of neural network with arrangement of single cerebral cortical neuron

Hayato Toriumi, Tomoyuki Kaneko (*LaRC,FB,Hosei Univ*)

Neural network model is necessary for understanding brain functions. Simple neural network model is needed to analyze the individual cell activity. In this study, we aimed to develop neural network with units consisted of single cerebral cortical neuron. We designed Agarose Micro Chamber (AMC) and Agarose Micro Channel (AMCn) that cell body adhered and neurites elongated respectively. We found the effect of AMC size on the elongation of neurite into AMCn. Accordingly, AMC diameter to construct neural network was set to 24~30 μm. We also made linear neural network composed with 4 units. These results suggested that cell body size was important factor for neurite elongation and AMC-AMCn structure contributed to construction of neural network model.

1Pos200 緑茶由来カテキンはナメクジの匂い嫌悪条件付け学習による記憶形成を強化する
Green tea-derived catechins enhance the long-term memory formation for an odor-aversive conditioning in the land slug

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We focus the effect of Epicatechin (EC) and Epigallocatechin gallate (EGCg), both flavanols found in many food (e.g. green tea and cocoa powder), on the present study. Here we investigated the effect of EC and EGCg on long-term memory formation of an odor aversive learning in the land slug, *Limax valentianus*. When slugs were preexposed to 15 μg/g solution of EC or EGCg 1h prior to the training, which typically results in memory lasting ~3 days, they formed LTM lasting at least 1 week. The procerebrum (PC), the center of odor processing, shows an oscillatory local field potential at about 0.7 Hz, which is modulated by conditioned odor stimuli. We also studied at the level of neural activities in the PC how EC or EGCg lead to the enhancing effects of the memory formation.

1Pos201 Sites for formation and storage of associative motor memory revealed by reversible expression of metabotropic glutamate receptor 1

Yasushi Kishimoto[1], Harumi Nakao[2], Kouichi Hashimoto[3], Kazuo Kitamura[4], Miwako Yamasaki[5], Kazuki Nakao[2], Masahiko Watanabe[5], Masanobu Kano[6], Atsu Aiba[2], Yutaka Kirino[1] ([1]*Kagawa. Sch. Pharm. Sci., Tokushima Bunri Univ.*, [2]*Cent. Dis. Biol. Integr. Med., Univ. Tokyo*, [3]*Sch. Med., Hiroshima Univ.*, [4]*Sch. Med., Univ. Yamanashi*, [5]*Sch. Med., Hokkaido Univ.*, [6]*Sch. Med., Univ. Tokyo*)

Eyeblink conditioning is a cerebellar dependent motor learning. However, where and how the memory is formed, stored and extinct has been a matter of long standing debate. To address this issue, we used mice bearing inducible and reversible expression of metabotropic glutamate receptor subtype 1 (mGluR1), specifically in Purkinje cells (PCs) of the cerebellar cortex. We show that mGluR1 in PCs is required for the formation of eyeblink memory but dispensable for the expression of once acquired memory. Establishment of persistent eyeblink memory requires mGluR1 in PCs and neuronal activity in the deep cerebellar nucleus. We conclude that associative motor memory is initially formed in the cerebellar cortex, transferred to the DCN.

1Pos202* 光センサー LOV2 ドメインの光反応中間体の構造揺らぎ検出
Time-resolved study on structural fluctuations of transient intermediates of the light sensor LOV2 domain

Shunrou Tokonami, Yusuke Nakasone, Masahide Terazima (*Dep. Chem., Sch. Sci., Kyoto Univ.*)

Phototropin is a blue light sensor in plants and algae. It contains two LOV domains as light sensing modules. It is well-known that the LOV2 domain shows a light-induced unfolding of its C-terminal helix, which is widely used for optogenetical tool. Here, applying the transient grating and the transient lens methods, we investigated the unfolding dynamics and structural fluctuations of transient intermediates. We observed significant changes in diffusion coefficient and partial molar volume associated with the unfolding. Analyzing the temperature dependence of the volume change, we detected an increase of the thermal expansion coefficient which strongly relates to the structural fluctuation. We discuss the importance of the fluctuation for protein reactions.

1Pos203* LOV ドメイン型光活性化アデニル酸シクラーゼ mPAC の光反応ダイナミクス
Photoreaction dynamics of LOV-domain-regulated photoactivated adenylate cyclase mPAC

Misato Ikoma, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci., Univ. Kyoto*)

Photoactivated adenylate cyclases (PACs) generate cAMP in a light-dependent manner. Interestingly, PACs possess different types of photoreceptor domains such as LOV, BLUF and GAF domains. In this study, we investigated the reaction dynamics of mPAC from cyanobacterium *Microcoleus chthonoplastes*. The mPAC contains the LOV domain as a light sensing module, and the photochemistry of its chromophore FMN is similar to those of other LOV domains. Using the transient grating method, we observed a significant change of diffusion coefficient upon photoexcitation, which is tentatively assigned to the conformational change of protein part. We compare the reaction with that of BLUF-regulated-PAC (OaPAC) to discuss the diversity of signaling mechanisms.

1Pos204* 光センサータンパク質 PYP と下流分子 PBP による励起波長依存的な会合・解離反応ダイナミクス
Excitation wavelength-dependent association and dissociation dynamics between light sensor protein PYP and its downstream protein PBP

Suhyang Kim[1], Yusuke Nakasone[1], Akira Takakado[2], Yoichi Yamazaki[3], Hironari Kamikubo[3], Masahide Terazima[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*Grad. Sch. Sci., Univ. Gakushuin*, [3]*Div. Mat. Sci., NAIST*)

PYP is a bacterial light sensor protein regulating a negative phototactic response. It contains a *p*-coumaric acid as a chromophore. Recently, its downstream partner protein PBP has been identified in *Rhodobacter capsulatus*, and the intermolecular interaction dynamics between PYP and PBP has been studied by the transient grating (TG) method. Since PYP has two absorption bands in UV and visible regions, we performed the TG measurements at UV and blue excitation. Interestingly, we found that the UV excitation leads to the complex formation with PBP, while the complex dissociates upon blue light excitation. The kinetics of the association and dissociation processes have also been investigated, and we will discuss the possibility that PYP plays a role of the color sensor.

1Pos205* TG 法を用いた OCP と FRP の時間分解相互作用ダイナミクスの検出
Detection of time-resolved interaction between OCP and FRP by using transient grating method

Takatoshi Ohata, Yusuke Nakasone, Masahide Terazima (*Grad. Sch. Sci., Univ. Kyoto*)

Orange Carotenoid Protein (OCP) is a blue light sensor in cyanobacteria, and has a biological function relating to the photoprotection. OCP contains a carotenoid molecule as a chromophore, and blue light irradiation causes a transition from the orange state (OCPO) to red state (OCPR). It is known that relaxation of OCPR to OCPO is accelerated by Fluorescence Recovery Protein (FRP). However, its molecular mechanism has been still unknown due to a lack of technique to detect the intermolecular process in time domain. In this study, we investigated the interaction dynamics between OCP and FRP using the transient grating (TG) method.TG signal of OCP is remarkably changed in the presence of FRP, which represents that the OCP associates the FRP in the light state.

1Pos206* 固体 NMR による KR2 のレチナール結合ポケットと Na+結合サイト間の水素結合ネットワークの構造解析
Structural analysis of hydrogen-bond networks between retinal binding pocket and Na+ binding site on KR2 by solid-state NMR

Rina Kaneko[1], Arisu Shigeta[2], Toshio Nagashima[3], Toshio Yamazaki[3], Keiichi Inoue[4,5], Hideki Kandori[5], Izuru Kawamura[1,2] ([1]*Grad. Sch. Eng. Sci., Yokohama National Univ.*, [2]*Grad. Sch. Eng., Yokohama National Univ.*, [3]*RIKEN*, [4]*Univ. Tokyo*, [5]*Nagoya Inst. Tech.*)

Krokinobacter rhodopsin 2 (KR2), a light-driven Na+ pump, has a characteristic protein-bound Na+ at the extracellular side. Our study of change in 15N NMR signal of protonated Schiff base suggested the presence of long-distance interactions between the retinal chromophore and extracellular Na+ binding site. Here, we investigated the alkali-metal ion dependent NMR chemical shift variations by solid-state NMR. Upon replacement of Na+ with Cs+, the several cross-peak shifts of the residues were observed and the structural differences in the extracellular side were found. Especially, the peak of Arg109 in the vicinity of retinal was significantly shifted. It is indicated that the Na+ binding site plays a role to regulate the hydrogen-bond networks in the resting state.

1Pos207 SEC-SAXS 法によるシロイヌナズナ由来フィトクロム B の構造解析
Structural analysis of Arabidopsis phytochrome B by small-angle X-ray scattering coupled with size-exclusion chromatography

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Phytochrome B (phyB) is a plant photoreceptor protein that regulates various photomorphogenic responses to optimize plant growth and development. PhyB, of which molecular weight is approximately 130 kDa, comprises an N-terminal photosensory module (PSM) and a C-terminal output module (OPM), and exists in two photoconvertible forms: a red-light absorbing (Pr) and a far-red light-absorbing (Pfr) form. Previous study revealed the crystal structure of PSM, but there are many unclear points including whole structure, domain arrangements, and mechanism of light-induced regulation. Here, we report the molecular shape of the Pr form Arabidopsis phyB and molecular properties of the Pfr form determined by small-angle X-ray scattering coupled with size-exclusion chromatography.

1Pos208 Structural basis of photo-stability of invertebrate rhodopsins

Midori Murakami (*Dept Physics, Nagoya Univ*)

In contrast to vertebrates, invertebrate metarhodopsins are thermally stable and hit back to the initial state by absorption of a second photon. To investigate the activation mechanism of invertebrate rhodopsins we have been performing crystallographic studies of squid rhodopsin. To date we have solved the structures of a series of photo-intermediates. Recently we obtained a new crystal of squid metarhodopsin and determined its structure. It reveals that helix architecture is in a closed form but with unlocked salt bridge of the DRY motif. We also found structural differences in the vicinity of the active site in comparison to bovine. Based on the results so far, we will discuss structural insights into the activation mechanism of bi-stable invertebrate rhodopsins.

1Pos209 ロドプシンの構成的活性変異体 M257Y のメカニズムに関する分子動力学シミュレーション
Molecular dynamics simulation study on the mechanism of constitutively active mutant M257Y of rhodopsin

Yuichiro Kanamori, Tadaomi Furuta, **Minoru Sakurai** (*Tokyo Tech*)

G protein-coupled receptors (GPCR) are seven transmembrane helix (TH) proteins that play an important role in the activation of intracellular G proteins. Constitutively active mutants (CAMs) are ubiquitously found among GPCRs and increase the inherent basal activity of the receptor. Here we focused upon a CAM M257Y of rhodopsin (Rh) and performed accelerated MD (aMD) simulation to elucidate the activation mechanism of this mutant. The aMD simulations indicated that in the mutant, K311 on TH8 approaches the ionic lock formed between R135 (on TM3) and E247 (on TM6). As a result, the ionic lock in the mutant becomes less stable than that in the wild type Rh, leading to an opening motion of TM6, which is essential for binding with the G protein.

1Pos210 ラマン光学活性で観るハロロドプシン多量体形成による活性部位の構造変化
Raman optical activity observes a clear structural change of active site caused by trimer formation of halorhodopsin

Shogo Ogawa[1], Tomotsumi Fujisawa[1], Takashi Kikukawa[2,3], Masashi Unno[1] ([1]*Grad. Sch. Sci. Eng., Saga Univ.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*, [3]*GSS, GI-CoRE, Hokkaido Univ.*)

Many microbial rhodopsins function in oligomer in the cell membrane, typically as light-driven ion pump. However, the correlation between the oligomer formation and the function is mostly unknown. A difficulty in studying the significance of the oligomerization is to detect the small but meaningful variations of proteins and their active sites caused by the quaternary structure change. In this study, we used Raman optical activity (ROA) spectroscopy, which is very sensitive to the molecular structure in solution, to analyze the chromophore conformational change in the oligomer formation of microbial rhodopsins. We carried out the ROA measurement of halorhodopsin from Natronomonas pharaonis to study the structural change of the active site in its trimer formation.

1Pos211 ラマン光学活性による光駆動型内向きプロトンポンプの研究
Near-IR Raman optical activity spectroscopy of inward proton pump rhodopsin

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A microbial rhodopsin with a novel function of light-driven "inward" proton pump activity was recently found from marine bacteria in deep sea. In the molecular mechanism of inward proton pumping, the conformation of the retinal chromophore can be an important factor that determines the direction of the proton transfer involving the active site. In this study, we used Raman optical activity (ROA) spectroscopy, which is sensitive to the three-dimensional structures of molecules, to analyze the conformation of the retinal chromophore in the inward proton pump. The ROA spectra showed the different signs between the inward proton pump and the conventional outward proton pumps. The chromophore conformation was determined using the quantum chemical calculations.

1Pos212 Actinotalea fementans 由来ヘリオロドプシンの物性解析
Molecular properties of Heliorhodopsin from Actinotalea fementans

Rei Abe-Yoshizumi, Ai Muto, Hideki Kandori (*Nagoya Inst. Tech.*)

Heliorhodopsin (HeR; 48C12) is a new family of rhodopsin, discovered in 2018. Slow photocycle suggests sensor function, whereas the interaction partner remains unknown. Functional analysis of HeR is not easy because of the difficulty to culture HeR-containing native cells. In this study, we identified the HeR gene from an actinomycete named *Actinotalea fermentans*, which can be cultured in the laboratory. The amino acid sequence of AfHeR is 57 % homologous to that of 48C12. To study their molecular properties, we constructed the expression system in *E. coli*. AfHeR has some unique molecular properties that are different from 48C12, such as unstability to pH and light. We will discuss molecular properties of HeR.

1Pos213 (6-4)光回復酵素の光反応過程における時間分解分光研究
Time-resolved spectroscopic study on photoreaction of (6-4) photolyase

Daichi Yamada[1], Takashi Nomura[1], Yuna Nakajima[2], Minoru Kubo[1] ([1]*Grad. Sch. Life Sci.*, [2]*Dept. Life Sci., Univ. Hyogo, Japan*)

(6-4) photolyases are DNA repair flavoenzymes that specifically revert UV-induced (6-4) photoproducts, into normal bases to maintain genetic integrity. So far, (6-4) photoproduct repair model via two photon processes has been proposed, but reaction intermediates remain unknown. For understanding the molecular mechanism, the DNA repair intermediates and the transient states of flavin in (6-4) photolyase have been necessary to determine by time-resolved UV-vis/ vibrational spectroscopy. In this study, we performed the time-resolved UV-vis to detect the intermediate upon one-photon illumination. In this presentation, we will discuss the repair mechanism in the (6-4) photolyase from the obtained experimental data.

1Pos214 集光クマリン色素を有する DNA 光回復酵素による高効率光駆動 DNA 修復
Enhanced light-driven DNA repair by DNA photolyase bearing light-harvesting coumarin chromophore

Yuma Terai[1], Ryuma Sato[2], Risa Matsumura[1], Shigenori Iwai[1], **Junpei Yamamoto**[1] ([1]*Grad. Sch. Eng. Sci., Osaka Univ.*, [2]*RIKEN BDR*)

Photolyases (PLs) catalyze light-driven DNA repair of carcinogenic UV-damaged DNA, via an electron transfer from the excited state of reduced flavin adenine dinucleotide, FADH-. The PLs are known to possess a secondary chromophore that absorbs light more than FADH- and transfers the energy to FADH- for enhancement of the DNA repair activity. Thus, the second chromophore is so-called a light-harvesting antenna pigment. So far, folates and flavin derivatives are known to function as the antenna. In this study, we newly found that a coumarin chromophore covalently-attached to thermophilic PL enhances its light-driven DNA repair activity. The efficiency highly depends on the amino acid residue where the chromophore is labeled, in agreement with the theoretical estimation.

1Pos215 Impact of a water molecule on photoreduction of (6-4) photolyase

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(6-4) photolyases ((6-4)PLs) are flavoproteins that can repair the ultraviolet-induced (6-4) photoproduct in DNA using blue light. To achieve the repair, their flavin adenine dinucleotide (FAD) cofactors need to be reduced to an FADH − form through a Trp chain in a light-dependent manner, called photoreduction. Previously, we showed that animal (6-4)PLs have four Trps involving photoreduction, and the fourth Trp is important to stabilize the radical pair generated during photoreduction. Although plant (6-4)PLs have only three Trps, they successfully perform photoreduction. In this study, we evaluated the effect of the water molecule close to the terminal Trp on photoreduction in a plant (6-4)PL, by combining the photoreduction assay and molecular dynamics simulation.

1Pos216 Characterization of Antarctic inward proton pumping microbial rhodopsins (AntRs)

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Microbial rhodopsins are membrane bound photoreceptors with seven known functions: ion-transporters (H+, Cl-, Na+), photosensors, channels (anions, cations), and enzyme rhodopsins. The retinal chromophore is bound in the middle of helix G, forming the retinal Schiff base (SB). We are characterizing a new group of inward H+ pumps which were isolated from a freshwater lake in Antarctica. They are missing a carboxylic acid adjacent to the SB; crucial to function in other proton pumps. AntR has light switchable metastable states exhibiting 13-cis, 15-syn retinal. The slow photocycle of AntR and robust transport is consistent with a double photon transport mechanism. Results of various visible and IR spectroscopic measurements on the WT and several mutants will be presented.

1Pos217 Inversion of Proton Transport Direction in Thermophilic Rhodopsin by Neutralizing the Secondary Counterion Asp229

Minori Kiyoshima[1], Takashi Kikukawa[2,3], Tomoyasu Aizawa[2,3], Makoto Demura[2,3], Yuki Sudo[4], Takashi Tsukamoto[2,3] ([1]*Grad. Sch. Life Sci. Hokkaido Univ.*, [2]*Fac. Adv. Life Sci. Hokkaido Univ.*, [3]*GSS, GI-CoRE*, [4]*Grad. Sch. Med. Dent. Pharm. Sci. Okayama Univ.*)

A microbial rhodopsin called thermophilic rhodopsin (TR) is a light-driven outward proton pump. Through mutational analysis of TR, we discovered that the neutralizing mutation for Asp229 (D229N), which is the secondary counterion of the protonated retinal Schiff base, inverted the proton transport direction. To understand its inward proton transport mechanism, we performed spectroscopic and electrochemical measurements. As a result, it was revealed that the photocycle of TR-D229N mutant was composed of K and P_{520}-intermediates, and the P_{520} was responsible for the inward proton transport accompanied with the deprotonation of Glu106. We conclude that the deprotonation of Asp229 is significant for precisely regulating the proton transport direction in the wild-type TR.

1Pos218 シゾロドプシンの内向きプロトン輸送経路の特性
Characterization of the inward proton transport pathway in Schizorhodopsin

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Schizorhodopsins (SzRs) were discovered in ASGARD archaea, which are the closest extant relatives of eukaryotes and in metagenomic scaffolds from putative bacteria. Multiple sequence alignments and phylogenetic analyses positioned SzRs as intermediates between type-1 rhodopsins and Heliorhodopsins. Measurement of light-dependent pH change showed that SzRs worked as light-driven inward proton-pumps in *E. coli* cells. To reveal the mechanism of inward proton-transport, we conducted site-directed mutational analysis of one of SzRs, AM_5S_00009. Some of the mutants for the residues expected to be located on the proton transport pathway abolished proton transport activities. Based on these results, we will discuss the mechanism of active inward proton-transport.

1Pos219* 紅色光合成細菌由来光捕集反応中心 1 複合体のスペクトル多様性と安定性におけるカルシウムイオンの役割
A dual role for calcium in expanding the spectral diversity and stability of LH1-RC photocomplexes of purple phototrophic bacteria

Michie Imanishi[1], Mizuki Takenouchi[2], Shinichi Takaichi[3], Shiori Nakagawa[4], Yoshitaka Saga[4], Shinji Takenaka[1], Michael Madigan. T[5], Jorg Overmann[6], Zheng-Yu Wang-Otomo[2], Yukihiro Kimura[1] ([1]*Grad. Sch. Agricultural Sci., Kobe Univ.*, [2]*Fac. of Sci., Ibaraki Univ.*, [3]*Fac. of Life Sci., Tokyo Univ. of Agriculture*, [4]*Dep. of Chem., Kindai Univ.*, [5]*Dep. of Microbiol.., Southern Illinois Univ.*, [6]*Microbiol., Braunschweig Univ. of Tech.*)

Purple photosynthetic bacterium Thiorhodovibrio (Trv.) strain 970 cells absorb the lowest energy among purple bacteria containing bacteriochlorophyll a, however, the cause remains unresolved. Here we characterized the light-harvesting 1 reaction center (LH1-RC) complex from Trv. strain 970 by spectroscopic and thermodynamic analyses. Our results clearly demonstrated that Trv. strain 970 integrates Ca2+ into its LH1-RC and extend its light-harvesting capacity. We discussed molecular mechanisms for the exceptional red-shifting of the LH1 absorption from Trv. strain 970 based on the known structural details of the Thermochromatium (Tch.) tepidum LH1-RC complex and proposed the dual role of Ca2+ in expanding the spectral diversity and stability of LH1-RC.

1Pos220 超分子複合体構造を構成する光合成アンテナ組成の解明
Elucidation of supramolecular components in photosynthetic antenna

Tetsuko Nakaniwa[1], Ryuichi Kano[2], Naoko Norioka[1], Soichiro Seki[2], Ritsuko Fujii[2,3], Genji Kurisu[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Sci., Osaka City Univ.*, [3]*OCARINA, Osaka City Univ.*)

As for the sunlight utilization, photosynthetic systems require to control the energy supply to the reaction center depending on the light intensities. Recently, the light-harvesting complexes (LHCs) from plant and green algae are considered to play a key role of the light control using dynamics of supramolecules associating the reaction center. LHC proteins form supramolecular, such as monomer or trimer, consisted of many isoforms. In plant LHC, it has been found that specific isoforms assemble specific supramolecules. In green algal LHC, the isoform composition in the supramolecules, however, is still not very clear. In this study, we established the method to analyze LHC components relating supramolecular formation as described in this poster.

1Pos221 Molecular mechanism of pH-dependent electron-flow regulation in photosystem II

Yuichiro Shimada[1], Seiryu Nakajima[1], Ryo Nagao[1,2], Takumi Noguchi[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*RIIS, Okayama Univ.*)

pH-dependent regulation of the relaxation of a reduced secondary quinone electron acceptor (Q_B^-) was recently proposed as a photoprotection mechanism of PSII (Nozawa *et al.*, 2018). A His residue near Q_B was predicted to be involved in this mechanism. Here, to identify the amino acid residue that is responsible for the pH-dependent electron-flow regulation in PSII, we characterized a D1-H252A mutant using delayed luminescence (DL). DL of WT showed faster relaxation of Q_B^- at higher pH as expected. In contrast, the H252A mutant showed no pH dependence in the relaxation rate, and the observed rate was similar to that of WT at a high pH. It was thus concluded that the protonation state of D1-H252 regulates the pH-dependent Q_B^- relaxation by shifting its redox potential.

1Pos222 緑藻の光捕集アンテナタンパク質 SCP の再構成
In-vitro reconstitution of light-harvesting complexes of a siphonous green alga, *Codium fragile*

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Siphonaxanthin (Sx) absorbs green region of sun light when bound to the photosynthetic light-harvesting pigment-protein complex called SCP in a green alga, Codium fragile. The red-shift of absorption in protein moiety may due to a specific interaction between Sx and amino acid residue, but precise structure has not clarified yet. In-vitro reconstitution is a powerful tool to figure out the specific interaction. The technique for plant light-harvesting complex has been established, but not for other pigment system. In this study, we examined the in-vitro reconstitution of SCP using inclusion body of recombinant SCP protein and a certain mixture of purified natural pigments. The reproducibility will be discussed in relation to the pigment compositions.

1Pos223 LH2 タンパク質の B800 部位へ再構成したクロロフィル誘導体のスペクトル特性変化に対するテトラピロール環構造の影響
Structural effects of chlorophyll pigments on their spectral properties induced by reconstitution into the B800 site in LH2 protein

Yoshitaka Saga, Madoka Yamashita, Kanji Miyagi (*Faculty of Science and Engineering, Kindai University*)

Exchange of cyclic tetrapyrroles, such as chlorophyll (Chl) and bacteriochlorophyll (BChl), in photosynthetic light-harvesting proteins is promising to regulate spectral coverage for photon capture and to improve the photosynthetic efficiency of these proteins. From this viewpoint, we examine insertion of various cyclic tetrapyrrole pigments into the binding site of B800 BChl a in light-harvesting complex 2 (LH2), which is a peripheral antenna protein in purple photosynthetic bacteria. Red-shifts of the Qy peak positions of 3-acetyl Chl a and 3-acetyl protoChl a, whose degrees of hydrogenation in the cyclic tetrapyrroles were different from BChl a, by insertion into the B800 binding site were smaller than that of BChl a.

1Pos224 ガラス基板表面におけるチラコイド膜の再構成と光合成機能解析
Reconstitution and functional analysis of thylakoid membrane on a glass substrate

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We reconstituted thylakoid membranes purified from spinach on a glass substrate as a continuous two-dimensional membrane. For facilitating the planer membrane formation, thylakoid membranes were mixed with phospholipid (DOPC) vesicles, and reconstituted into the scaffold of patterned polymeric bilayer. The electron transfer activity of PSII was confirmed by the changes in chlorophyll fluorescence as the electron acceptor or inhibitor were added. We observed the generation of NADPH on the surface of thylakoid membranes, confirming the electron transfer activity of PSI. These results support the feasibility of using reconstituted thylakoid membranes as the experimental platform to evaluate the molecular machinery of photosynthesis.

1Pos225 光化学系複合体と酸化グラフェンを用いた水素発生
Hydrogen production using photosystem and graphene oxide

Shunsuke Sone[1], Mriko Miyachi[2], Shota Tanaka[1], Hisataka Ohta[1], Yoshinori Yamanoi[2], Akihide Iwase[1], Akihiko Kudo[1], Hiroshi Nishihara[2], Tatsuya Tomo[1] ([1]*Tokyo University of science*, [2]*The University of Tokyo*)

Photosynthetic organisms convert solar energy into chemical energy. The quantum yield of initial reaction of photosystem is very high by the forces of natural selections. Graphene oxide (GO) is good material for its remarkable electronic properties. Exploiting this photovoltaic abilities of photosystem (PS) and excellent electronic properties of GO for biohybrid device is one of the key research themes for sustainable energy. In this study, we generated PS I-GO-PS II conjugates dispersed in a solution aimed at application in artificial photosynthesis. Using this system, hydrogen production proved that the photoexcited electron originated from PSI and PS II with light irradiation. This conjugate will be a useful bio-device for artificial photosynthesis.

1Pos226 緑藻ミル糸状体のカロテノイド蓄積における培養時光条件の影響
The effect of different light regimes for carotenoid accumulation of a macro green algae, Codium fragile, in filamentous form

Soichiro Seki (*Osaka city university , department of Chemistry, Research institute for natural science and technology*)

Chloroplast generally accumulate β-carotene, lutein (Lu), violaxanthin (Vx), and 9'-cis neoxanthin as major carotenoids. Recently, we have reported that a green alga, Codium species accumulate neoxanthin in all-trans conformation when cultivate under high light conditions. It also has a unique carotenoid composition: it accumulates siphonaxanthin and α-carotene as substitute for Lu and β-carotene, respectively, and few Vx. However, the detail of carotenoid biosynthesis and accumulation profile are not clarified yet. In this study, we investigated precise carotenoid accumulation profiles of Codium fragile cultured in filamentous form under different color/intensity light conditions.

1Pos227 Time-resolved infrared analysis of proton release pathways in photosynthetic water oxidation using a D1-N298A mutant and NO_3^- substitution

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Photosynthetic water oxidation is performed at the Mn_4CaO_5 cluster through a cycle of intermediates called S states. Although proton release has been detected in the S_0->S_1, S_2->S_3 and S_3->S_0 transitions, pathways for individual protons remain to be identified. In this study, we investigated the proton release pathway in the S_2->S_3 transition by time-resolved infrared measurements of a D1-N298A mutant, which perturbs a channel near Y_Z, in combination with NO_3^- substitution for Cl^-, which perturbs a channel near Cl^-. It was shown that the S_2->S_3 transition was significantly retarded upon D1-N298A mutation, whereas further retardation was not observed by NO_3^- substitution. This result suggests that Y_Z channel is mainly used for proton release in the S_2->S_3 transition.

1Pos228 QM/MM analysis of the protonation structure of the S_0 state in the water-oxidizing Mn_4CaO_5 cluster

Masao Yamamoto, Shin Nakamura, Takumi Noguchi (*Grad. Sch. Sci., Nagoya Univ.*)

Photosynthetic water oxidation is performed at the Mn_4CaO_5 cluster in PSII. Proton release has been detected in the S_0-S_1 transition, and either O4 or O5 in the Mn_4CaO_5 cluster has been proposed as the site of proton release. However, the protonation structure of the S_0 state remains unclarified. In this study, we calculated the vibrations of carboxylate ligands to the Mn_4CaO_5 cluster using the QM/MM method to identify the protonation site in the S_0 state. An S_1-minus-S_0 infrared spectrum in the symmetric COO^- region calculated using an O4H model of S_0 showed a better agreement with the experimental spectrum than that using an O5H model. It is thus suggested that O4 is protonated in the S_0 state and this proton is released upon S_1 formation through the O4-water chain.

1Pos229 Infrared microspectroscopic study on water oxidation in a single photosystem II microcrystal

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Microcrystals of PSII have been used in time-resolved x-ray crystallographic studies to clarify the mechanism of water oxidation. It is crucial to know whether the reactions efficiently proceed in PSII microcrystals. In this work, we used FTIR microspectroscopy to analyze water oxidation reactions in a single PSII microcrystal. FTIR difference spectra of a single microcrystal measured by a transmission method showed features virtually identical to the spectra of PSII in solution as well as the spectra of numerous PSII microcrystals previously obtained with an ATR method that reflects the reactions near the crystal surfaces. This result indicates that water oxidation proceeds with relatively high efficiencies even in the inside of a PSII microcrystal.

1Pos230 光化学系 II の表在性タンパク質による水分解 Mn_4CaO_5 クラスターの S_2 構造異性体平衡の制御機構
Equilibrium of the S_2-state isomers of the water-oxidizing Mn_4CaO_5 cluster in photosystem II regulated by extrinsic proteins

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The S_2 state of the water-oxidizing Mn_4CaO_5 cluster in photosystem II (PSII) of green plants has two isomers with open and closed cubane conformations, which show $g = 2.0$ multiline and $g = 4.1$ EPR signals, respectively. In cyanobacterial PSII, however, the $g = 4.1$ signal is usually not observed. Here, we examined the effects of extrinsic proteins on the equilibrium of the two isomers in red algal and plant PSII. The EPR spectrum of PSII complexes from a red alga *C. merolae* showed only the $g = 2.0$ multiline signal. Removal of the extrinsic proteins did not affect this spectrum, whereas it eliminated the $g = 4.1$ signal in spinach PSII. These results indicate that binding of the extrinsic proteins in green plant PSII stabilizes the closed cubane form in the S_2 state.

1Pos231 Dynamics of photosystem II protein complexes as observed by high speed atomic force microscopy

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The molecular structure and photosynthetic mechanism of photosystem II (PSII), which has a function of water oxidation, have been extensively studied using various methods. However, the large-scale dynamics of PSII protein complexes have never been reported. Here we employed high-speed atomic force microscopy (HS-AFM) to directly observe the dynamics of PSII. HS-AFM observation showed the dissociation of extrinsic proteins and subsequent repetitive conformational changes in the PSII proteins. Although these structural changes were not directly related to photoreactions, it appeared to be triggered by destruction of the Mn cluster, the catalytic center of water oxidation. This result suggests that the Mn cluster stabilizes the local conformations of the PSII proteins.

1Pos232* 酵素型ロドプシン（Rh—PDE）の非対称的 pH 効果
Asymmetric pH effect on the enzyme rhodopsin, Rh-PDE

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The enzyme rhodopsins are membrane proteins composed from a rhodopsin domain which binds all-*trans* retinal as a chromophore and an enzyme domain at the C-terminal cytoplasmic side. In this study, we focus on a molecule that functions as light-activated phosphodiesterase (Rh-PDE). Our research purpose is to elucidate activation mechanism accompanied by light absorption. Here, we studied pH dependence of the enzyme activity of Rh-PDE. Interestingly, the pH dependency of the enzymatic activity toward cAMP and cGMP hydrolysis showed asymmetry, in which cAMP hydrolysis was elevated whereas cGMP hydrolysis was reduced by acidic pH. The substrate selectivity of Rh-PDE is discussed based on the present results and structural information.

1Pos233* 光活性型 bZIP モジュールであるフォトジッパーにおける Gln317 の役割
The role of Gln317 in a light-activatable bZIP module, Photozipper

Itsuki Kobayashi, Osamu Hisatomi (*Grad. Sch. Sci., Univ. Osaka*)

Photozipper (PZ) is an engineered protein containing a basic region/leucine zipper (bZIP) domain and a light-oxygen-voltage-sensing (LOV) domain of Aureochrome-1. We have reported that blue light irradiation induced the dimerization of PZ and increased its affinity for the target DNA. In LOV domain, a Gln residue closely locates at FMN is highly conserved. To elucidate the role of this Gln residue (corresponding to Q317 of PZ), we prepared recombinant proteins in which Q317 was replaced to Val, Leu, Asn, Ser and Glu. Interestingly, these mutants showed higher affinities for the target DNA than PZ in the dark state with similar affinities of PZ in the light state. Our data suggested the Q317 plays a role on the stabilization of monomeric form in the dark state.

1Pos234* プロトンポンプ型ロドプシンによる緑藻クラミドモナスの 非光化学的消光(NPQ)の人為的光制御
Optical control of non-photochemical quenching (NPQ) in the alga *Chlamydomonas reinhardtii* by light-driven proton pump rhodopsins

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Microbial rhodopsins have been used as a fundamental tool for optogenetics, a method to control biological activities by light in animals. Here, as a model for optogenetics in plants, we employed the alga *Chlamydomonas reinhardtii* to control non-photochemical quenching, NPQ. NPQ is a protection system from excess solar energy, which is triggered by acidification of the lumen. We generated transgenic alga expressing inward and outward H^+ pump rhodopsins in the thylakoid membranes. The algae expressing inward and outward H^+ pumps showed 2-fold larger and slightly lower NPQ capability, respectively, than the control, suggesting the successful control of NPQ. Together with the physiological effect of light on the alga, we will discuss the prospect of optogenetics in plants.

1Pos235* Theoretical study on molecular mechanics of natural anion channelrhodopsin GtACR1

Takafumi Shikakura, Cheng Cheng, Shigehiko Hayashi (*Kyoto Univ.*)

Channelrhodopsins are photo-sensitive channel proteins with a retinal chromophore and are utilized in optogenetics. Recently a structure of an anion-conducting channelrhodopsin, GtACR1, was determined by X-ray crystallography. We theoretically examined protonation states of counter ion groups (Glu68 and Asp 234) and distributions of water molecules and a Cl- anion in a putative channel which are key properties for photo-sensitive ion channel conduction. Ab initio QM/MM free energy geometry optimizations revealed tight coupling of the protonation states and distributions of water molecules and a Cl- anion, providing a molecular insight into electrostatic environment in the anion conducting channel.

1Pos236 蝶のカモフラージュや擬態模様にみる多要素構造
Multi-component systems of camouflage and mimicry in butterfly wing patterns

Takao Suzuki (*NARO*)

Modular design is a fundamental principle in living systems such as gene regulatory networks and developmental cellular mechanisms. Well then, how about morphological structures? It was a long-standing conundrum in evolutionary developmental biology. Recently, our team investigated animal eyes and various butterfly patterns of camouflage and mimicry and revealed that these can be decomposed into an assembly of multiple components. In summary, I proposed a conceptual framework: multicomponent systems, where complex traits are decomposable into an assembly of subcomponents (reducibility) and build up by various combinatorial arrangements of such components (composability). Our studies have begun to reveal a combinatorial building logic of morphological structures.

1Pos237 巨大化大腸菌の再生過程可視化
Regeneration of Escherichia coli giant protoplasts

Kazuhito Tabata, Takao Sogo, Yoshiki Moriizumi, Hiroyuki Noji (*Department of Applied Chemistry, The University of Tokyo*)

The spheroplasts and protoplasts of cell wall-deficient (CWD) bacteria are able to revert to their original cellular morphologies through regeneration of their cell walls. However, whether this is true for giant protoplasts (GP), which can be as large as 10 μm in diameter, is unknown. we prepared GP from E. coli and showed that they can return to E. coli's original morphology, and that they are capable of colony formation. Microscopic investigation revealed that the regeneration process took place through a variety of morphological pathways. We also report the relationship between GP division and GP volume. These results indicate that E. coli is a highly robust organism that can regenerate its original form from an irregular state such as GP.

1Pos238 大腸菌を用いた実験室内進化におけるタンパク質の配列進化速度の制約
Constraint of protein evolution speed in de novo experimental evolution of *E. coli*

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Constraints of the rate of protein sequence evolution have been a central question in evolutionary biology. Numerous correlates of the protein evolutionary rate have been reported previously, which triggers explorations of underlying constraints in the rate of protein sequence evolution. Is there any universal constraint governing the rate of protein sequence evolution? Here, we analyzed genomic mutations of Escherichia coli accumulated in de novo evolution under an artificial condition. We detected the negative correlation between the rate of protein sequence evolution and gene expression level. The negative correlation retained after controlling for non-random mutation rate, suggesting a constraint by purifying selection rather than any constraint of mutation rate.

1Pos239 (1SEA-5) Molecular Dynamics of Nucleosome Assembly

Giovanni Brandani, Shoji Takada, Cheng Tan (*Dept Biophysics, Div Biology, Grad School Science, Kyoto University*)

The dynamic assembly of nucleosomes underlies the regulation of epigenetic patterns throughout genomes. We employed molecular dynamics simulations to extensively characterize the environmental factors modulating the kinetics of nucleosome assembly. Markov state modeling reveals the existence of a rich set of metastable partially assembled nucleosome structures, and that specific DNA bending motifs facilitate assembly by directing the binding of histones toward their target. We further show how the flexibility of nucleosomal DNA across yeast promoters is related to both nucleosome occupancy and gene expression, highlighting that sequence can directly contribute to chromatin organization by either facilitating or inhibiting nucleosome assembly at specific genomic regions.

1Pos240 全原子 Motion Tree による側鎖運動の記述とドメイン運動との連動
Full-atom Motion Tree detects side-chain motions and their coupling with domain motions

Ryotaro Koike, Motonori Ota (*Grad. Sch. Info., Nagoya Univ.*)

Proteins are flexible molecules and change the structures by external stimuli, e.g. ligand binding. The comparison of structures in ligand-bound and -unbound forms reveals the structural changes. We developed a computational tool, Motion Tree, to compare protein structures and describe the structural change using Cα atoms. Motion Tree identifies various motions in proteins from small loop motions to large domain motions. We have extended the tool to employ all heavy atoms and describe motions in a finer-resolution. Motion Tree of full-atom version was applied to 88 proteins exhibiting a simple domain motion. A large number of side-chain motions were detected. It is revealed that several side-chain motions tend to be coupled with large movement of domain.

1Pos241 マルチカノニカル法を用いた蛋白質球状ドメイン外の相互作用の解析
Analysis of the protein-protein interaction between regions external to globular domains with multi-canonical molecular method

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Elucidation of the principles of protein-protein interactions is a central issue in biophysics. In particular, the nature of interactions between regions external to globular domains such as linker and loop were unclear. In this study, we defined such region, i.e., interacting two regions without non-local contacts as “floating interaction segment (FIS)”. We focused on four FISs retrieved consisting of about 10 residues from the PDB and extracted these FISs from their entire structures. The stability of interactions between pairs of 10 residue peptides (FISs) were analyzed by multi-canonical molecular dynamics (McMD) simulation. As a result, although these FISs stably kept interactions, the FISs didn’t fold a specific structure and yielded multi-modal conformations.

1Pos242 並列タンパク質間相互作用予測システム MEGADOCK の高速化・仮想化
Acceleration and virtualization of parallel protein-protein interaction prediction system MEGADOCK

Masahito Ohue[1], Hiroki Watanabe[1,2], Kento Aoyama[1,2], Yutaka Akiyama[1] ([1]*Sch Computing, Tokyo Tech*, [2]*RWBC-OIL, AIST*)

Protein-protein interaction (PPI) plays a core role in cellular functions. We developed an ultra-high-throughput PPI prediction system based on rigid-body docking, named MEGADOCK. Here we show that acceleration and virtualization of MEGADOCK. We selected Docker which is an implementation of Linux containers, and implemented a distributed computing environment of MEGADOCK with virtual machines (VMs) on Microsoft Azure, and evaluated its performance. Both when MEGADOCK directly performed on VM and when it performed with Docker containers of MEGADOCK on VM, execution speed achieved almost equal even if #cores was increased up to ~500 cores. Container techniques have large contributions to improve productivity and reproducibility of scientific researches.

1Pos243 蛋白質における Non-local 接触を持たない領域に関する統計解析
Segments without non-local contacts in protein structures

Kota Kasahara[1], Shintaro Minami[2], Yasunori Aizawa[3], Ryohei Kondo[4], Takuya Shimato[4], Takuya Takahashi[1] ([1]*Coll. Life. Sci., Ristumeikan Univ.*, [2]*EXCELLS, NINS*, [3]*Sch. Life Sci., Tech., TokyoTech*, [4]*Grad. Sch. Life Sci., Ritsumeikan Univ.*)

Three-dimensional structure of protein is primarily established by a tight network of intramolecular contacts. However, regions without intramolecular non-local contacts exist in proteins, where non-local contact means the contact between amino acid residues separated with many residues along the primary sequence. In this study, we performed statistical analyses of protein segments without non-local contacts, termed “floating segments”, based on the protein data bank (PDB). As a result, 0.72 % of residues were in floating segments. Floating segments are enriched in intermolecular interfaces with polypeptides and polynucleotides. In terms of amino acid propensity, floating segments preferred Gly and Ala, and disfavored Arg and His.

1Pos244 Direct coupling analysis of amino-acid sequences based on the Hopfield-Potts model

Kai Shimagaki, Martin Weigt (*Sorbonne Universite, Paris-IV*)

Direct coupling analysis (DCA) is a statistical mechanics-inspired approach, which models amino-acid sequences via Potts models. The couplings of these models are used in protein-structure prediction, the energies to predict mutational effects in proteins. Conventional DCA uses a fully connected model, each amino-acid in each position can interact with each other, thus resulting in a huge number of parameters to be inferred from limited sequence data. Here, we introduce a method based on Hopfield-Potts model, which largely reducing parameter number. Furthermore, the Hopfield patterns are interpretable in terms of sequence motifs.

1Pos245 繊維状粒子凝集の CA タイプ解析
Attempts at CA-type formal analysis of fibrous assembly of particles

Takashi Konno (*Biomath.Med.Univ.Fukui*)

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.

1Pos246 エピジェネティックな状態変化が細胞のがん化に及ぼす影響のランドスケープ理論による解析
Landscape analyses of epigenetic state change in cancerization

Yutaro Kameyama, Masaki Sasai (*Dept. Appl. Phys., Grad. Sch. Eng., Univ. Nagoya*)

Recurrence and metastasis of cancer cells make complete cure of cancer diseases difficult. It was suggested that cancer stem cells(CSCs) are responsible for the recurrence and metastasis. Therefore, analysis of the pathway for generating CSCs is important. We developed a model of transitions among CSCs, stem cells, differentiated cells and cancer cells by extending the gene network model of Li and Wang(Cancer Res 2015) to include epigenetic degrees of freedom such as histone modification and DNA methylation patterns. In this extended model, each cell state is represented as a basin in the landscape of gene expression level, and the landscape structure is significantly affected by both regulatory interactions among genes and epigenetic state kinetics in individual genes.

1Pos247 筋分化過程で誘導される細胞競合は IGF シグナルを同期化する
Synchronization of IGF signal by cell competition during myogenesis

Fumihiko Hakuno, Masato Masuda, Ryosuke Okino, Shin-Ichiro Takahashi (*Dep. App. Ani. Sci., Grad. Sch. Agri. Life. Sci., The Univ. of Tokyo.*)

Cell competition serves as a fitness sensing mechanism in which less fit cells are eliminated from the cell layer by the fitter cells. We recently have shown that cell competition could be monitored during myogenesis by the differential level of IRS-1 protein, which is a major mediator of IGF-I signal. Based on these observations, cell competition was simulated during myogenesis using cellular automaton. IRS-1 protein level was calculated in each cell by simulating IGF signal. Our simulation results indicated that cell competition events synchronized the IGF-I signal with neighbor cells. Taken together with the other experimental data, we concluded that cell competition-induced IGF-I signal synchronization is required for cell fusion to form multinucleated myotubes.

1Pos248 モジホコリ変形体における輸送管ネットワークの分岐則
Direct observation of branching rules in transportation network of *Physarum* plasmodium

Masahiro Shibata, Atsuko Takamatsu (*Dept. of Elec., Eng. & Biosci., Waseda Univ.*)

Murray's law is a well-known branching rule in biological transportation networks. Murray proposed that flow rate in transportation tubes is proportion to the cube of the radius when energy consumption is minimized. This results in the branching rule with branching exponent three: the cube of the radius of a parent tube equals the sum of the cubes of the radii of the daughters'. To verify whether the branching rule holds for plasmodium of *Physarum polycephalum*, consisting of tubes transporting protoplasm. The relation between flow rate and tube radius was investigated. The results suggest that flow rate is proportion to the cube of the radius in the branches of the mature tubes, whereas the branching exponent is lower than three in the immature branches.

1Pos249 シグナル伝達分子の細胞膜上クラスター形成機構の数理研究
Mathematical study on cluster formation of signaling proteins on the cell membrane

Hiroaki Takagi (*Dep. Phys., Sch. Med., Nara Med. Univ.*)

In order to reveal spatial distributions of signaling proteins on the cell membrane and their functional significance, we have performed spatial statistical analysis of cAR1(GPCR) positional data in Dictyostelium cells detected by PALM microscopy. Spatial density of cAR1 within 100nm scale is higher than that of homogeneous Poisson distribution, and the cluster size of cAR1 shows power-law like distribution. Here, to elucidate the mechanism of such spatial distributions of receptors, we mathematically studied several models of cluster formations and their extensions in spatial point process theory. We discuss the possible relationship between the spatial distribution of receptors and the spatial distribution of membrane lipids as their "fields".

1Pos250 線虫の graded ニューロンはどのようにして確率的な 2 状態スイッチングダイナミクスを生成するか？
How do graded neurons generate stochastic binary switching dynamics in C. elegans?

Yuishi Iwasaki (*Fac. Eng., Ibaraki Univ.*)

In the whole-brain imaging data of C. elegans [Toyoshima et al, 2016], some neurons show different responses (nearly in anti-phase) to the same stimuli. The different neural responses can be explained by stochastic binary switching of the neurons. Unlike in mammals, neurons show not action potentials but graded membrane potentials in C. elegans. I present how do the graded neurons generate stochastic binary switching dynamics. In this work, a stochastic differential equation is used to simulate neural activity of model neurons. In addition, Fokker-Planck equation is used to calculate the time evolution of the probability density function of the stochastic neural activity. I discuss about the noise intensity dependence on change of the probability density function.

1Pos251* Effects of in vivo rhythm-damping mutations to KaiA on circadian rhythm in vitro

Masahiro Wakayama[1], Risa Imada[1], Yuki Nakamoto[1], Rie Kumagai[1], Keisuke Serizawa[2], Masahiro Ishiura[3], Kousuke Maki[1] ([1]*Grad. Sch. Sci., Nagoya Univ.*, [2]*Sch. Sci., Nagoya Univ.*, [3]*Nagoya Univ.*)

Clock proteins KaiA, KaiB and KaiC are responsible for circadian rhythm of cyanobacterium Synechococcus elongatus PCC7942 not only in vivo but also in vitro. KaiA and KaiB regulate KaiC phosphorylation level through formation of complexes. Previous study reported that several KaiA mutations damped the circadian rhythm in gene expression (Uzumaki et al., 2004). However, the effects of these KaiA mutations on KaiC phosphorylation rhythm remain unknown. To address this question, we measured KaiC phosphorylation reaction and circadian reaction with these KaiA mutations including F224S, H271A, C273Y. As the result of the measuring, we found that F224S and C273Y affect only circadian reaction while H271A affects both KaiC phosphorylation reaction and circadian reaction.

1Pos252* Exploring a simply phosphorylation cycle by using phosphorylation site variants of clock protein KaiC

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Clock proteins, KaiA, KaiB and KaiC, exhibits circadian rhythm in the presence of ATP in vitro. Among the proteins, KaiC has two phosphorylation sites Ser431(S) and Thr432(T), whose phosphorylation state oscillates in the order of ST->SpT->pSpT->pST until going back to ST. Phosphorylation at S431 is expected to be more difficult than that at T432 because of an additional introduction of like charge(p) to negatively charged pT at the adjacent site. This leads to an idea that a fraction of SpT may go back to ST instead of proceeding to pSpT. Here we designed a “simple” phosphorylation cycle interconverting between ST and SpT by using KaiC variants and adjusting experimental conditions. It suggests existence of such a “simple” cycle in natural Kai circadian reactions.

1Pos253 (1SHA-4) 高圧力下で早くなるシアノバクテリアの概日周期
(1SHA-4) Pressure accelerates the circadian clock of cyanobacteria

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The oscillation period lengths of circadian clocks in diverse organisms are mostly independent of ambient temperature, known as temperature compensation. However, nothing has been known about pressure effects on circadian clocks. Here we report pressure effects on the cyanobacterial circadian clock, consisting of KaiA, KaiB and KaiC proteins. We found that the cycle of the KaiC phosphorylation was accelerated from 22 hours at 1 bar to 14 hours at 200 bar. We also found that ATPase activity measured in kcat of KaiC at 200 bar was 1.5 times greater than that at 1 bar. These results match the proposed correlation between the circadian period length and the ATPase activity of KaiC period-mutant proteins.

1Pos254 心筋細胞ネットワークにおける局所伝導ゆらぎの幾何学的理解
Geometrical understanding of the local fluctuation in propagation of excitation conduction in cardiomyocyte network

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To understand the cell number dependence for stabilization of the excitation conduction of cardiomyocyte networks, we investigated the relationship between the local and the global fluctuations in propagation of excitation conductions. We measured the extracellular field potentials and excitation conduction time of cardiomyocyte cells in the line patterned networks arranged on a multi electrode array. The results showed that the fluctuation of excitation conduction stabilized as the conduction distance increased and the excitation conduction for long range distance was sum-up of local fluctuations. Those results indicate that the minimum conduction distance can stabilize the excitation conduction of the cardiomyocyte network.

1Pos255 Observation of direction-dependent asymmetrical propagation velocities in excitation conduction in a same cardiomyocyte networks on a chip

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To elucidate the importance of geometric arrangement and community effect of cells for creating the quasi in vivo assay, we investigated the propagation manner of action potential conductions in a series of different cardiomyocyte networks. We analyzed extracellular field potential propagations of cardiomyocytes in those two-dimensional networks, which were formed on a multi-electrode-array chip with an agarose microfabrication technology. We found the propagation velocity was asymmetrical in forward and reverse excitation conduction at the same areas in those networks. The results suggest the simple cardiomyocyte network has direction-dependent anisotropic propagation of conduction in a same network even though each single cardiomyocyte is isotropic.

1Pos256 水素化アモルファスシリコンと紫外可視光変換で増強されたセンサのためのゲル電気化学素子と分子薄膜
Gel electrochemical element and molecular film for sensor enhanced by hydrogenated amorphous silicon and ultra violet light conversion

Koki Shimanaka[1], Shota Murakami[1], Kairi Shimazaki[1], Kishiro Seino[1], Hikaru Hatakeyama[1], Shu Mugita[1], Hiroshi Masumoto[2], Takashi Goto[3], Yutaka Tsujiuchi[1] ([1]*Mat Sci Akita Univ*, [2]*FRIS Tohoku Univ*, [3]*IMR Tohoku Univ*)

Photo-controlled film system, by using ionic conduction in laminated gels, ultra violet visible light conversion system, and the effect of hydrogenated amorphous silicon film, have been studied for the purpose of fabrication of functional biosensor. Firstly Langmuir Blodgett films of different fluorescent molecules were fabricated. And an emission spectrum analysis of films was conducted. As the result ultra violet visible light conversion system functions as the detection of sample properties. Secondly rectification properties in gel electrochemical element were influenced and induced to be controllable by light irradiation. Thirdly, the effects of hydrogenated amorphous silicon film on the sensor samples are discussed.

1Pos257 On-chip differential analysis of sequential phagocytosis on identical position of single macrophages

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We investigated the hysteresis of phagocytosis in macrophages with comparing the phagocytic response time differences during sequential stimulations of antigens (IgG coated polystyrene beads) on identical position of isolated macrophages using optical tweezers and a mechanical manipulator. Before reaching to the maximum beads number of phagocytoses (50 beads for 2 μm, 14 for 4.5 μm, 7 for 6 μm, 3 for 20 μm), the engulfment in early intervals showed similar phenomenon with no hysteresis regardless of antigen size, whereas the last a few phagocytosis increased significantly caused by the prolongation of the first recognition step and the second engulfment phase maintained constant, indicating the saturation of phagocytosis influence to their antigen recognition.

1Pos258 水素化アモルファスシリコンに積層した脂質とバクテリオロドプシン複合膜の構造変化観察
Observation of structural change of lipid film and lipid and bacteriorhodopsin film laminated on hydrogenated amorphous silicon film

Hikaru Hatakeyama[1], Hiroshi Masumoto[2], Takashi Goto[3], Yutaka Tsujiuchi[1] ([1]*MatSci AkitaUNIV*, [2]*FRIS TohokuUNIV*, [3]*IMR TohokuUNIV*)

A molecular film contains bacteriorhodopsin (BR) and lipid film on retinoic acid film on hydrogenated amorphous silicon (a-Si:H) as the substrate and a film that contains BR film directly fabricated on a-Si:H were analized by FTIR spectroscopy and compared with each other. FTIR reflection absorption spectrums of DMPC LB film above fatty acid film. DFM image of RetA3L+DMPC2L film and a structural model are discussed. DFM image obtained by measurement from an angle of 45, or, 90 degrees upon magnification indicates gradually protruding curved surface portions with a maximum height difference of 9 nm and width of approximately 200nm. Further, observations of structural change of composite film of lipid and bacteriorhodopsin indicate a new aspect of biomolecule.

1Pos259 高速走査レーザーマイクロダイセクションシステムの開発
Development of a high-speed scanning laser microdissection system

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Laser microdissection (LMD) is a microscope-based method for precise separation of biological specimens by scanning a focused laser beam. LMD enables non-contact handling of RNA, DNA, and protein to study their functions in specific cells in living tissues. In common LMD systems, two prisms with heavy inertial weight are used to deflect a laser beam, which takes a long time to separate a specimen and thus can cause heat damage to target biomolecules. Therefore, in this study, we have developed a new LMD system into which a high-speed driven mirror was incorporated. The LMD system increased the scanning speed by 100 times compared with conventional systems.

1Pos260 光-電子相関顕微鏡法（CLEM）による同一試料観察に向けた相関・位置合わせ精度の評価
Evaluation of correlation and alignment accuracy toward the same sample observation by CLEM

Yuki Gomibuchi[1], Risa Ezoe[2], Hiroko Takazaki[1], Takuo Yasunaga[1] ([1]*Dept. of Phys. Info. Tech., Kyushu Inst. Tech.*, [2]*Dept. of Biosci. Bioinfo., Kyushu Inst. Tech*)

Characteristics of the LM and the EM. To watch the same sample and the same area, correlative light and electron microscopy (CLEM) is useful. Here, for alignment, we used fluorescent beads as a marker, with performing coordinate calculation and stage movement by SerialEM and so evaluated their errors. Then, when the targets were located within the marker group within a square of 90 μm, the coordinate errors were less than 5 μm. Under these conditions, the target within 10 μm could be observed in an estimated area. At present, alignment evaluation was performed using beads of different colors and sizes. Thus, we will follow real specimens on the grid with markers to verify alignment accuracy.

1Pos261 原子間力顕微鏡にひと工夫 ー*Volvox* 1 個体の推進力を「直接」測るー
A trick to atomic force microscopy enabling direct measurement of forces generated by swimming *Volvox* spheroids

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Here we report a new experimental system using modified scanning probe microscopy to directly measure forces of swimming microorganisms. A cantilever is deflected upon collision of swimming microorganisms. The force generated by a single microorganism can be calculated from the vertical displacement and the spring constant of the cantilever. To evaluate the capability of this system, we measured forces generated by swimming in two *Volvox* species, *V. rousseletii* (~5000 cells) and *V. carteri* (~2000 cells) and found that their forces were 16.2 ± 9.0 nN and 6.6 ±3.6 nN, respectively, which well agrees with the estimation by Stokes law. This is the first demonstration of direct force measurement generated by swimming microorganisms.

1Pos262 Intron seqFISH enables transcriptome-wide visualization of genome organization and nascent transcription in single cells

Yodai Takei[1], Sheel Shah[2], Wen Zhou[1], Eric Lubeck[3], Jina Yun[1], Chee-Huat Linus Eng[1], Noushin Koulena[1], Christopher Cronin[1], Christoph Karp[1], Eric Liaw[2], Mina Amin[4], Long Cai[1] ([1]*California Institute of Technology*, [2]*University of California, Los Angeles*, [3]*Stanford University*, [4]*University of California, Riverside*)

Recent single-cell studies have revealed significant heterogeneity at the levels of transcription and chromosome organization in individual cells. Visualization of the transcriptome and the nuclear organization in situ has been challenging for single-cell analysis in order to directly examine the interplay between those modalities. Here, we present a multiplexed single-molecule in situ imaging method, intron sequential fluorescence in situ hybridization (intron seqFISH) that allows imaging of up to 10,421 genes at their nascent transcription active sites in single mammalian cells, combined with multiple rounds of single-molecule mRNA seqFISH and immunofluorescence. This work opens up imaging-based discovery-driven studies of biological processes in many contexts.

1Pos263* (3SEA-2) ラマンイメージングを用いた細胞内の水の可視化とラベルフリー細胞内温度測定への応用
(3SEA-2) Raman imaging of water in a cell and its application to label-free evaluation of intracellular temperature

Toshiki Sugimura, Shinji Kajimoto, Takakazu Nakabayashi (*Grad. Sch. Pham. Sci., Tohoku. Univ*)

We have previously reported quantitative evaluation of water in a cell using Raman microscopy. Water density in nucleus was shown to be higher than that in cytoplasm in a HeLa cell. Here, we applied Raman imaging of water to perform label-free evaluation of intracellular temperature.We measured Raman images of HeLa cells and evaluated the intensity ratio of two regions on the O-H band at different temperatures. The intensity ratio changed linearly with temperature, and the calibration line was obtained both in nucleus and in cytoplasm. We measured the increase in cytoplasm temperature with the addition of FCCP in a cell. A rise in the intracellular temperature was observed, indicating that intracellular can be measured using Raman images of water without staining dye.

1Pos264* Elastin 様ポリペプチドに基づく分子温度センサー
Molecular Thermometer Based on Elastin-Like Polypeptide

Cong Vu, Tetsuichi Wazawa, Takeharu Nagai (*ISIR, Osaka Univ.*)

Microscopic visualization of temperature dynamics in cells is a promising method to explore cell-physiological activities. Development of thermometers based on polymers or proteins has been attempted. However, improvement in sensitivity of thermometers is needed for tracking fast temperature changes in live cells. Here, we report a highly-sensitive ratiometric thermometer contained temperature sensing domain of elastin-like polypeptide (ELP) fused with a CFP and a YFP. At elevated temperatures, the structure of ELP shrinks leading to FRET from CFP to YFP. Detectability to temperature was the highest (20%/ °C) among existing thermometers. We will demonstrate applicability of this thermometer to visualize temperature changes during cellular processes and drug treatments.

1Pos265* 新規小分子プローブによるアクチン繊維の可視化と光操作
Visualization and manipulation of actin cytoskeleton by using novel small molecular probes

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Actin filament (F-actin) is one of the major components of cytoskeleton in eukaryotic cells and provides cells with mechanical support and driving force for movement. Herein, we show that HMRef, a rhodol fluorescent dye, can clearly visualize actin cytoskeleton without any washing or transfection treatment. HMRef can be applied for various cell lines or primary cultured cells including those SiR-actin can't be applied for. We also utilized HMRef as a binding scaffold for F-actin and developed a CALI probe named 'GLIFin'. GLIFin can induce fragmentation of F-actin upon laser irradiation and decrease migration rate only one part of epithelial cell sheets. We expect these probes would be potentially practical for discoveries about biophysics or morphogenesis.

1Pos266 (1SCP-6) グルタミン酸受容体を介した植物の長距離 Ca^{2+} シグナル
(1SCP-6) Long-distance Ca^{2+} transmission via glutamate receptor channels in plants

Masatsugu Toyota[1,2] ([1]*Dept Biochem and Mol Biol, Saitama Univ*, [2]*University of Wisconsin-Madison*)

In the vertebrate central nervous system, glutamate acts as an excitatory neurotransmitter, regulating synaptic transmission via activation of glutamate receptor channels and facilitating long-distance information exchange throughout their bodies. Similarly, plants sense local stresses, such as insect attack, and transmit this information throughout their bodies to rapidly activate defence responses in undamaged organs. Here we show that glutamate is a wound signal in plants that leaks from damaged cells and activates the *GLUTAMATE RECEPTOR LIKE* family of ion channels in *Arabidopsis*, resulting in the intracellular Ca^{2+} transmission to distant organs (Toyota et al Science 2018). These data help understand long-range molecular signaling networks in animals and plants.

1Pos267 (1SDP-4) Visualization and quantification of biological samples by high-speed atomic force microscope

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High-speed atomic force microscope (HS-AFM) allows us to visualize the dynamic structural changes of biological samples and to intuitively understand their functional mechanism under physiological conditions at high spatiotemporal resolution. Using HS-AFM, recently, not only the direct observation of dynamic conformational change at the single-molecule level but also the quantification of the binding/dissociation process between individual molecules has been carried out. Additionally, we are trying to extend the range of objects by combining with fluorescence microscopy or by obtaining an additional information such as mechanical properties of samples. In this presentation, we will show analytical method of several experiments and discuss about the capability of HS-AFM.

1Pos268 (1SGA-8) 細胞内動態をサブセルレベルで制御する温和な NanoHeating 技術
(1SGA-8) A Thermodynamic Tool for Mechanobiology Research: Mild Nanoheating Technology to Alter Subcellular Dynamics

Satoshi Arai[1], Nandus Ferdi[2] ([1]*Res. Inst. Sci. Eng., Waseda Univ.*, [2]*WABIOS*)

Recent years, we developed the nanoheating technology that enables to heat up the intracellular local place using a photothermal dye-based nanoparticle with a near infrared laser. Previously, we demonstrated our technology in the induction of apoptosis in cancer cells under harsh elevated temperature. Here, we report several applications using more "mild" heating to change cellular activities. For example, we showed that mild heating could induce the skeletal muscle contraction based on the alteration of protein-protein interaction. In the other case, we found that mild heating affected the dynamics of intracellular ATP. We do believe our methodology will be helpful to provide the viewpoint of thermodynamics to mechanobiology research.

1Pos269 クライオ電子顕微鏡により明らかになったノロウイルスの動的構造変化
Dynamic Structural Change of Norovirus Revealed by Cryo-electron Microscopy

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Human norovirus (HuNoV) is the major cause of epidemic nonbacterial gastroenteritis. Since the mechanism of infection has not been well studied, there is no efficient vaccines and remedies yet. In this study, we produced murine noroviruses (MNVs) by reverse genetics as a surrogate of HuNoV, and investigated their capsid structures by single-particle cryo-electron microscopy. As a result, protruding (P) domains of MNVs were stabilized by interactions between adjacent P domains. Furthermore, MNVs showed two types of conformations by rotation of P domain. The infectivity rate to cells in the two conformations was different, and the one conformation showed higher accessibility of the cellular receptor CD300lf causing greater viral infectivity.

1Pos270 化学発光ビリルビンセンサーの開発
Development of bioluminescent unconjugated bilirubin indicator

Yukino Ito[1], Yoshiyuki Arai[2], Mitsuru Hattori[2], Takeharu Nagai[2] ([1]*Graduate School of Frontier Biosciences, Osaka University*, [2]*The Institute of Scientific and Industrial Research, Osaka University*)

Unconjugated bilirubin (UC-BR) is a metabolite of hemoglobin, and its concentration in blood works as a common index for several diseases. High concentration of UC-BR causes newborn jaundice and encephalopahy. Here, we report a ratiometric bioluminescent indicator that is composed of luciferase and UnaG, UC-BR binding green fluorescent protein (Kumagai et al., Cell, 2013). When UC-BR binds to UnaG, FRET occurs from luciferase to UnaG resulting in emission color shift. By comparing this color change under various conditions, we demonstrated detection of UC-BR in newborn blood by a spectrophotometer. This indicator would help not only make blood-based BR diagnosis easy, but also understand heme metabolism by in vivo imaging.

1Pos271 高速原子間力顕微鏡 1 分子計測データを用いた粒子フィルタ法によるリンカー DNA 付きヌクレオソームの動的構造解析
Dynamic structure analysis of nucleosome with linker DNAs by particle filter method using single molecule measurement data by HS-AFM

Sotaro Fuchigami[1,2], Toru Niina[1], Shoji Takada[1,2] ([1]*Grad. Sch. of Science, Kyoto Univ.*, [2]*CREST, JST*)

High-speed atomic force microscopy (HS-AFM) is a powerful technique to directly observe biomolecules in dynamic action. However, its spatiotemporal resolution is not enough to reveal their molecular details. In the present study, we have developed a four-dimensional structure analysis method of single molecule measurement data by HS-AFM combined with coarse-grained molecular simulation using data assimilation with particle filter. As a target biomolecule, we selected a nucleosome with linker DNAs,. We performed particle filter simulation using synthetic AFM images as observation data, and confirmed that this combined method works well. We also performed the simulation at different ionic strengths, suggesting the ability to estimate its value in experiment.

1Pos272 化学発光トロンビンセンサーの開発
Development of chemiluminescent thrombin sensor toward whole body imaging of living mice

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Thrombin is one of the proteases which play a central role in blood coagulation. Increasing thrombin activity generates thrombosis, and it causes a myocardial infarction and some disease. Measurement of thrombin activity in our body is significant for not only therapy but also understanding mechanism of thrombosis. In this research, we developed a BRET-based sensor for thrombin activity, which is composed of a thrombin recognition peptide, a luciferase and a fluorescent protein. With this sensor, we succeeded to detect thrombin activity in mouse blood by detection of emission color change, which was induced by cleavage of the thrombin recognition peptide in the sensor by active thrombin. In future, we will observe the distribution of thrombin activity in mouse body.

1Pos273 ESPT 型バイオセンサーの設計
Design of fluorescent biosensors based on Excited State Proton Transfer (ESPT) in the chromophore of a fluorescent protein

Kazunori Sugiura[1,2], Toru Hisabori[2], Shoko Mihara[2], Takeharu Nagai[1] ([1]*ISIR, Univ. Osaka*, [2]*CLS, Tokyo Tech*)

GFP has two absorption peaks at around 400nm and 480 nm, corresponding to a neutral and ionized form of the chromophore, respectively. On the other hand, the fluorescence emission is only single peak at 510 nm from ionized chromophore. This is owing to the excited state proton transfer (ESPT) from neutral form of excited chromophore to Glu222, producing ionized form of excited chromophore. If the ESPT could be inhibited, GFP with a neutral form of excited chromophore should blue emission. Therefore, this could be used to design an emission ratiometric indicator. We will demonstrate a rational design of a redox indicator capable of emission ratiometry, and discuss the superiority of this design in comparison with another one such as FRET-based indicators.

1Pos274 4 次元透過型電子顕微鏡：理論とシミュレーション
4-Dimensional Transmission Electron Microscopy: Theory and Simulation

Kuniaki Nagayama (*N-EM Labos LLC*)

We will report on 4D electron microscopy of a transmission type (4D-TEM). 4D microscopy, irrespective to TEM or STEM, is a non-interferometric method relying on wave fields defined on 4D coordinates of the position(2D) and the direction(2D) and yields differential imaging by tracing the deflection of electrons individually incident to the particular position of objects. As shown last year, 4D reciprocity theorem guarantees the equivalence of 4D-STEM and 4D-TEM but practically several advantages may rest with 4D-TEM, which enables a huge sensitivity enhancement to improve the spatial resolution for biological molecules such as proteins and DNAs. Theoretical prediction for the sensitivity enhancement is to be verified with an EM simulation software developed for 4D-TEM.

1Pos275 環境の温度変化に対する細胞応答の分子機構
The molecular mechanism of cell response to environmental temperature change

Hiroki Shibata[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ of Tokyo*, [2]*PRESTO, JST*)

Environmental temperature has a great impact on life. Living things have adapted to temperature change in the course of evolution by acquiring temperature-responsive systems including those at cellular level. However, the mechanism of response to physiological temperature change inside cells has not been revealed. Here we show that physiological temperature change affects intracellular mRNA dynamics. By culturing COS7 cells at moderately low temperature, we found that the diffusion coefficient of mRNA decreased non-linearly and mRNAs were localized around the nucleus. Furthermore, artificial heating in these cells resulted in the suppressed temperature increase. These results suggested that cells can adapt to environmental temperature change by altering RNA dynamics.

1Pos276 相平衡状態にある核小体内領域における核小体構成タンパク質の 1 分子動態と超解像分子局在解析
Single-molecule dynamics and localization of nucleolar proteins in phase-separated compartments of nucleolus

Supanut Sirisukhodom[1], Daiki Matsumoto[1], Yuma Ito[1], Noriko Saitoh[2], Kumiko Sakata-Sogawa[3], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Tech*, [2]*Dept. of Cancer Biol., The Cancer Inst. JFCR.*, [3]*Grad. Sch. Agr. Sci., Tohoku Univ.*)

Nucleolus maintains its structure by phase separation without being surrounded by biological membranes, and is divided into different compartments: FC, DFC and GC. We examined the physical properties using simultaneous super-resolution and single-molecule imaging of constituent proteins in these compartments. Single-molecule analysis in the interlayer DFC region showed two types of dynamics: fast diffusion across the outermost GC region and slow diffusion in the interaction state. Super-resolution analysis visualized the DFC as several clusters with a diameter of 500 nm to 1 μm. The analyses in the GC showed distinctly different molecular dynamics and localization. The result suggests that the ribosomal protein supply is performed in the vicinity of DFC.

1Pos277 Development of designable RNA-binding proteins for visualization and manipulation of authentic RNAs in living cells

Akira Takai[1], Yasushi Okada[1,2] ([1]*BDR, RIKEN*, [2]*Univ. of Tokyo, Grad. Sch. of Sci., Dept. of Phys.*)

In this study, we report the development of designable RNA-binding protein (dRBP), which is programmable to bind to the RNA of interest. We first established an ELISA-like assay using our bright bioluminescent protein, Nano-lantern (Takai et al., PNAS 2015), and showed our dRBPs have high affinity and specificity to the target RNAs. We also showed our dRBPs can be used for the visualization of the authentic RNAs including Actb mRNA or lncRNA Neat1_2 in living cells. Furthermore, manipulation of the localization of the Actb mRNA using the dRBP fused to constitutively active kinesin resulted in the elongation of cellular processes. These data suggest that our dRBP would serve as a powerful tool for visualization and manipulation of authentic RNAs in living cells.

1Pos278 相分離に関わるヘテロクロマチンタンパク質 HP1α の 1 分子超解像イメージングによる分子局在と動態
Dynamics and localization of Heterochromatin protein 1α involved in phase separation using single-molecule and super-resolution imaging

Takahiro Maeda[1], Yuma Ito[1], Shin-Ya Isobe[2], Chikashi Obuse[2], Makio Tokunaga[1] ([1]*Sch. Life Sci. Tech., Tokyo Tech*, [2]*Biosci. Grad Sch Sci., Osaka Univ*)

Heterochromatin protein 1 α (HP1α) regulates gene expression by causing chromatin compaction. This is supported by phosphorylation of HP1α N-terminal extension, which promotes the formation of liquid-liquid phase separation. We examined the roles of HP1α in regard to the phase separation in simultaneous single-molecule and super-resolution microscopy using phase-separation inhibitor, histone methyltransferase inhibitor and HP1α mutants. Single-molecule tracking analysis correlated with nanometer localization revealed features of HP1α dynamics both dependent and independent of the phase separation and of HP1α dimerization. We will discuss the role of HP1α in chromatin dynamics with respect to the phase separation and interactions with other proteins.

1Pos279 Imaging transcriptional dynamics of the endogenous gene with a bright fluorogenic RNA

Tetsuro Ariyoshi[1], Yasushi Okada[1,2] ([1]*RIKEN BDR, Cell Polarity Regulation*, [2]*Dept. Phys., Grad. Sch. Sci., UTokyo*)

Recently we have developed a bright fluorogenic RNA, Romanesco, that enables quantitative visualization of mRNA in living cells. Romanesco has been applied to study spatiotemporal dynamics of exogenously expressed mRNA, however, visualizing endogenously expressed low-copy mRNA has been hampered by high background signals. Here we report new methods to remove the background signals efficiently by taking advantage of the photochemical properties of Romanesco. These new imaging methods enabled us to extract the Romanesco fluorescence signal selectively, resulting in a more than 20-fold increase in signal-to-background ratio. Using these methods, we succeeded in visualizing endogenous mRNA expression and revealed the frequency responses of the gene transcriptional dynamics.

1Pos280 ネクロプトーシスに伴う DAMPs 放出の LCI-S による可視化
Live Cell Imaging for Secretion Activity (LCI-S) of DAMPs Release Accompanying with Necroptosis

Yoshitaka Shirasaki[1,2], Mai Yamagishi[1], Sotaro Uemura[1] ([1]*Dept. of Biological Sciences, Grad School of Science, The Univ. of Tokyo*, [2]*JST PRESTO*)

We developed a platform allowing to track the secretion dynamics of a single cell, Live Cell Imaging for Secretion activity (LCI-S) applied it to the DAMPs release during necroptosis. Necroptotic L929 cells were found to exhibit HMGB1 release in two modes: burst and sustained mode (Nat Commun, 2018, 9, 4457). CHMP4B knockdown impaired sustained mode, suggesting membrane dynamics modulated the release. Besides, A20 KO mice exhibit IL-1-dependent arthritis caused by necroptosis of myeloid cells, but the relationship between IL-1 production and necroptosis are unclear. LCI-S on A20-KO BMDM revealed IL-1 release during necroptosis and no neighbouring cell exhibited effective cell death before it, indicating necroptotic cells were the IL-1 source (Nat Cell Biol, 2019).

1Pos281* 単一分子伝導計測に基づく表面上の DNA ハイブリダイゼーションの反応速度論解析
Kinetic investigation of DNA hybridization on surface using single-molecule conductance measurement

Takanori Harashima, Yuki Jono, Tomoaki Nishino (*Sch. Sci., TokyoTech.*)

We investigated single-molecule kinetics of DNA hybridization based on electrical conductance measurements of DNA by scanning tunneling microscopy (STM). The conductance increases upon DNA hybridization because the STM tip and substrate was bridged by the resulting duplex. We found that the hybridization efficiency on a metal surface is strongly regulated by the interaction between the metal surface and the DNA base. Moreover, the kinetic analysis of the (de)hybridization processes indicated the presence of the partially hybridized DNA as the intermediate state, which affects the hybridization efficiency on a surface. This research provides an unprecedent way to investigate kinetic processes of biological molecules at single-molecular scale on a surface.

1Pos282* 制御・情報技術の統合による集団内細胞行動特徴の定量解析
Quantitative analysis of collective cell migration by integration of controlled in vitro experiment and information processing

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The mathematical model could provide a powerful tool for elucidating the mechanism of complex system in a multicellular organization, but various mathematical models have been proposed for a single tissue formation because there is no evaluation. Here, we have achieved to establish the system of assessing the validity of those mathematical models quantitatively. Controlling the initial conditions in vitro can reduce the experimental noise to improve the reproducibility of the developed pattern formation. Then, feature extraction of reproducible data of collective cells converts a large amount of data into the meaningful information. By comparing these features with in silico results repetitively, the mathematical model can be validated and optimized.

1Pos283* 分散培養心筋細胞と心臓組織片の電気生理学的信号の同期
Synchronization of electrophysiological signal between dispersed cardiomyocytes and cardiac tissue piece

Toru Nakamura, Chiho Nihei, Tomoyuki Kaneko (*Laboratory for Reconstructive Cell biology, Department of Frontier Bioscience, Hosei University, Grad. School of Science and Engineering*)

Heart disease is the top cause of death in the world. Transplantation of cardiomyocytes sheet has been developed for treatment of ischemic heart disease. However, beating synchronization mechanism of dispersed cardiomyocytes and cardiac tissue piece is still unclear after transplantation of cardiomyocytes sheet. To analyze these signal synchronization process, we measured these extracellular potential by Multi Electrode Array (MEA) system. As a result, one of stable beating rate became a pacemaker regardless of dispersed cardiomyocytes or cardiac tissue pieces. Therefore, it was suggested that the stability of beating was an important factor for signal synchronization. It was assumed that the cardiomyocytes sheet with a stable beating rate could induce the arrhythmia.

1Pos284 DNA ナノデバイスを制御する DNA 生成回路の検証
Characterization of DNA Generation Circuits for Controlling DNA Nanodevices

Ken Komiya, Teruya Enomoto, Masayuki Yamamura (*Sch. Comp., Tokyo Tech.*)

We developed various DNA generation circuits that generate single-stranded DNA strands as signals for directing operation of DNA nanodevices. Their modular architecture allows permutation of DNA generation [1] and rapid DNA amplification at a constant “low” temperature (L-TEAM reaction) suitable for DNA computing and molecular diagnosis [2, 3]. In the present study, we experimentally investigated the characteristics of DNA generation circuits for achieving the sophisticated systems function of DNA-based artificial molecular systems.
[1] New Generation Computing, 2015, Vol. 33, No. 3, pp. 213-229
[2] Org Biomol Chem, 2019, DOI: 10.1039/c9ob00521h
[3] Anal Bioanal Chem, 2019, DOI: 10.1007/s00216-019-01878-z

1Pos285 Photo-control of Ras nucleotide exchange reaction using the inhibitor peptides modified with spiropyran derivative

Kenichi Taii[1], Nobuyuki Nishibe[1], Kei Sadakane[2], Shinsaku Maruta[1,2] ([1]*Dept. of Bioinfo., Grad. Sch. of Eng., Soka Univ.*, [2]*Dept. of Sus. Inno., Fac. of Sci. and Eng., Soka Univ.*)

Ras belongs to a family of the small G-proteins. Recently, peptide inhibitors of Ras GDP-GTP exchange were found. Photochromic molecule, spiropyran isomerize between hydrophilic SP and hydrophobic MC forms photo-reversibly upon visible and UV lights. It is known that zwitterion of spiropyran MC isomer forms dimer and induce secondary structural change of peptide modified with spiropyran. In this study, we designed and synthesized Ras peptide inhibitors based on interface with GEF which contains two cysteine residues. They were modified with SP-maleimide (SP-MA) stoichiometrically. We studied the conformational changes of the peptide accompanied by photoisomerization of spiropyran and photoreversible inhibition of GDP-GTP exchange of Ras.

1Pos286 アポフェリチンを使ったマグネタイト単結晶ナノ粒子の作製
Synthesis of single crystal magnetite nanoparticles in apoferritin cavity

Tomoko Kanamaru, Daisuke Katayama, Naoki Takasihima, Takeshi Narusima, **Hideyuki Yoshimura** (*Dpt. Phys., Meiji University*)

To get homogeneous nanoparticles (NPs), protein (apoferritin) cavity has been utilized as a reaction chamber. Apoferritin is an iron storage protein found in many biological species, known to mineralize several metal ions in vitro. It is a hollow, spherical protein composed of 24 subunits, H-subunit which has Fe(II) oxidation site and L-subunit without it. Here we report synthesis of magnetite (Fe3O4) nanoparticle with single crystal domain. Recombinant with H-subunit has Fe(II) oxidation site and thus oxidation occurs very quickly at each oxidation site in the cavity. In this reason, synthesized nanoparticles appear as amorphous or polycrystalline. To realize directed crystal growth, we controlled the number of H-subunit in an apoferritin molecule.

1Pos287 (1SCA-5) 光からエネルギーを合成しタンパク質合成をする人工光合成細胞の構築
(1SCA-5) Artificial photosynthetic cell producing energy for protein synthesis

Samuel Berhanu[2], Takuya Ueda[3], **Yutetsu Kuruma**[1] ([1]*JAMSTEC*, [2]*ELSI, Titech*, [3]*Grad. Sch. of Front. Sci., Univ. of Tokyo*)

Construction of an artificial cell widens our understanding of living organisms, but the mechanism to synthesize its own constituents by self-sufficient energy has not been developed so far. Here, we constructed an artificial cell that produces ATP by light and synthesizes its own constituent proteins. The artificial cell contains a photoreactive organelle which artificially consists of ATP synthase and bacteriorhodopsin. The photo-synthesized ATP eventually drove the synthesis of the components of the organelle, resulted in enhancement of the organelle activity through the positive feedback of the products. Our artificial photosynthetic cell system paves the way to construct an energetically independent artificial cell.

1Pos288 人工細胞-生細胞ハイブリッドバイオシステムの創成
Synthesis of artificial/living cell hybrid biosystems

Masamune Morita, Kaoru Katoh, Naohiro Noda (*Biomed. Res. Inst. (BMRI), AIST*)

An important issue of “bottom-up synthetic biology” is the design and creation of vesicle-based artificial cells. The biological cells are forming in complex structures such as having organelles inside. The organelles are thought to be formed by symbiosis with bacteria. In order to test this hypothesis, we trying to create an artificial/living cell hybrid biosystems that encapsulating bacteria into vesicles and investigate their functions. This presentation describes the current state and the development of a hybrid biosystem project; a new variety of artificial cells that can be developed by combining cellular and synthetic parts in a single vesicle. This hybrid biosystem will pave the way toward an advanced model protocell, which mimics evolution.

1Pos289 *Mycolicibacterium smegmatis* のストラクトーム解析
Structome analysis of *Mycolicibacterium smegmatis*

Hiroyuki Yamada[1], Masashi Yamaguchi[2] ([1]*Res. Inst. Tuberculosis., JATA.*, [2]*Mycol. Res. Cent., Chiba*)

Mycolicibacterium smegmatis, basonym *Mycobacterium smegmatis*, was performed. As *M. smegmatis* has often been used in molecular biological experiments and experimental tuberculosis as a substitute of highly pathogenic *Mycobacterium tuberculosis*, it has been a task to compare two species in the same genus, *Mycobacterium*, by structome analysis. 220 serial ultrathin sections were examined by TEM and the cell profiles were measured. All measurements in *M. smegmatis*, except cell length, are significantly higher than those of *M. tuberculosis*. In addition, these data may explain the more rapid growth of *M. smegmatis* than *M. tuberculosis* and contribute to the understanding of their structural properties, which are substantially different from *M. tuberculosis*.

1Pos290 Identification of lipid interactions in the transmembrane regions of human Na+, K+-ATPase

Dhani Ram Mahato, Magnus Andersson (*Dept. Che., Ume Univ.*)

The function of ion transporting Na+,K+-ATPase (NKA) depends on the surrounding lipid environment of plasma membrane. Certain lipid types activate different confirmations and known to stabilize the structural confirmation induced by the interaction of lipids at different sites named as site A, B and C within the transmembrane domains. An atomic scale MD simulation approach was used to identify the interacting lipids within the transmembrane regions and their specific sites of interaction. Two states of NKA, E1 and E2 were homology modeled and embedded into a artificial plasma membrane. The site C, which contains disease causing mutants at the lipid-protein interface, interacts with the POPE lipids.

2Pos001* Mycoplasma mobile のモーターを構成するタンパク質 MMOB1620 の構造解析
Structural analysis of MMOB1620 which composes Mycoplasma mobile's motor

Hiroki Sato[1], Aya Kodama[2], Hisashi Kudo[3], Koji Ooka[4], Syunji Suetaka[3], Yuuki Hayashi[3], Munehito Arai[3,4], Makoto Miyata[1,2] ([1]*Graduate School of Science, Osaka City University*, [2]*Faculty of Science, Osaka City University*, [3]*Dept. Life Sci., Univ. Tokyo*, [4]*Dept. Phys., Univ. Tokyo*)

Mycoplasma mobile, a pathogenic bacterium of fish glides with a unique mechanism. The force is generated from a protein complex which is thought to have evolved from F-type ATPase. The structure of this protein complex has been clarified by cryo electron microscopy at a resolution of 6.4 Å, and the protein components are partially assigned on the structure. Then, we focused on an unassigned protein component, MMOB1620 composed of 293 amino acid residues. The circular dichroism of a recombinant protein was consistent with the contents of secondary structure predicted from the amino acid sequence. The limited proteolysis showed that the region of 1 - 76th amino acid residues is flexible. Structural analyses based on small angle X-ray scattering (SAXS) are underway.

2Pos002* 異なる長さの C 末端領域を持つテロメア繰り返し配列結合タンパク質 AtTRP1 の DNA 結合領域に対する構造研究
Structural studies for DNA binding domain of telomere repeat binding protein, AtTRP-1 with different size of C-terminal region

Shunta Kojima[1], Hayato Morita[2] ([1]*Grad. Sch. Sci., Josai.Univ*, [2]*Fac.Sci., Josai.Univ*)

Telomere is indispensable to hold the gene structure during chromosome replication process. AtTRP is one of the telomere-binding protein found in *Arabidopsis thaliana*. This protein contains a Myb like DNA binding domain and resembles with several initiator-binding proteins found in higher-plants. In our previous study, we found that Myb-like DNA binding domain, alone, does not have the ability to bind with telomere repeat sequence, and the presence of C-terminal flexible region is needed. In this study, we have overexpressed several types of ^{15}N labelled AtTRP-1 Myb-like domain with different length of C-terminal region and studied the relationships between the structure and the binding affinity of Myb-like domain.

2Pos003* PaCS-MD によるタンパク質–タンパク質複合体の解離シミュレーション
Protein-protein complexes dissociation simulated by Parallel Cascade Selection Molecular Dynamics

Yoshiki Miyazawa[1], Duy Phuoc Tran[2], Kazuhiro Takemura[2], Akio Kitao[2] ([1]*Grad. Sch. LST.,Tokyo Tech*, [2]*Sch. LST.,Tokyo Tech*)

Simulating association and dissociation processes of protein-protein complexes are important for understanding life phenomena on the molecular basis. However, conventional molecular dynamics (MD) cannot capture these rare events. Parallel Cascade Selection Molecular Dynamics (PaCS-MD) is one of the techniques to induce the conformational transitions from one state to another without applying bias, and has been successfully applied to dissociation simulation of protein-ligand complexes. In this work, we used PaCS-MD to simulate dissociation process of protein-protein complexes. By applying Markov State Modeling to the generated trajectories, we were able to reproduce experimental values of binding free energy of the protein complexes.

2Pos004 Deep-Autoencoder に基づいたホモロジーモデリングソフトウェアの開発
Development of Deep-Autoencoder based Homology Modeling software

Masaya Furue, Mitsutaka Nemoto, Lisa Matsukura, Naoyuki Miyashita (*BOST KINDAI Univ.*)

Currently, the much primary sequence of the protein has been elucidated by genome analysis using next-generation sequencers and so on. However, though the tertiary structure usually has been solved by the NMR spectroscopy, X-ray structure analysis, and cryo-EM, the number of the known tertiary structures of proteins are less. Thus, the structure prediction or structure modeling for protein have been used to obtain the tertiary structure of proteins. In this study, we developed deep-autoencoder based homology modeling method for soluble protein (mDeepHoMe : MD based Deep Learning for Homology Modeling). We used the trajectories of Molecular Dynamics simulations of template proteins as learning data. We will show the examples for modeling using this program.

2Pos005 Caged-GTP を用いたがん遺伝子産物 Ras の SACLA, SPring-8, NMR による GTP 加水分解過程の構造変化の解明
Structural changes on GTP hydrolysis of oncogene product Ras revealed by SACLA, SPring-8 and NMR using photo-controllable caged-GTP

Yoshiteru Makino[1], Takashi Kawamura[2], Shigeyuki Matsumoto[1], Eriko Nango[3], So Iwata[3], Takashi Kumasaka[2], Fumi Shima[4] ([1]*Grad. Sch. Med., Kobe. Univ.*, [2]*Protein Cryst. Anal. Div., JASRI.*, [3]*Grad. Sch. Med., Kyoto Univ.*, [4]*Grad. Sch. Sci. Tec. Innov., Kobe Univ.*)

The ras oncogene products Ras is a small GTPase which functions as a molecular switch by cycling between GTP-bound active and GDP-bound inactive forms. GTP-bound Ras adopts two interconverting conformational states, inactive State 1 and active State 2. In order to reveal the structural dynamics of Ras in state transition and GTP hydrolysis process, we demonstrated time-resolved structural analysis of photo-controllable caged-GTP-bound Ras by SACLA, SPring-8 and NMR. After photo-irradiation, we succeeded in the first observation of natural GTP-bound Ras by SACLA and Spring-8. Further, the result of time-resolved measurements by NMR, Spring-8 and SACLA revealed novel information on the structural dynamics of Ras for developing potent Ras inhibitors.

2Pos006 アブラナ科植物の自家不和合性を制御するタンパク質 SRK/SP11 複合体の Rosetta と accelerated MD を用いた構造モデリング
Computational modeling of SRK/SP11 protein complexes using Rosetta and accelerated MD simulations

Yoshitaka Moriwaki[1], Tohru Terada[1,2], Koji Murase[1], Kentaro Shimizu[1] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

S-locus receptor kinase (SRK) in pistil and a small ligand protein, *S*-locus protein 11 (SP11) in pollen are responsible for *Brassicaceae* self-incompatibility (SI). If both proteins are derived from the same *S*-locus, the pollen will be rejected to prevent self-pollination. Although two haplotypes of their complex crystal structures have been determined, there remains more than 25 haplotypes with unknown structure. Here we report that most of the SRK and SP11 structures were successfully generated by using Rosetta combined with accelerated MD simulations. Moreover, some modeled SRK/SP11 complexes showed high affinity in the calculations. Our modeling method contributes to an understanding of the self-recognition toward artificial control.

2Pos007 粗視化シミュレーションによる CDK4 の構造変化に関する研究
Conformational transition of CDK4 by using coarse-grained simulations

Kazutomo Kawaguchi, Hidemi Nagao (*Inst. Sci. Eng., Kanazawa Univ.*)

Cyclin-dependent kinases (CDKs) are attractive targets for therapeutics against cancer. Association of CDK4 to cyclin D is one of the triggers to express genes required for the early stage of cell cycle. CDK4 is a client protein activated by association with heat shock protein 90 (HSP90). We have studied the activation mechanism of HSP90 from the viewpoint of the ligand binding. In this study, we focus on the conformational transition of CDK4 and investigate the conformational stability of native and open states of CDK4 with/without cyclin D3 by using our coarse-grained model. We observed an intermediate state of CDK4 stabilized by cyclin D3 during the conformational transition.

2Pos008 Single particle analysis of silkworm lipid transfer protein complex, lipophorin

Shunsuke Kita[1], Kazuhiro Mio[2], Mika Hirose[3], Kenji Iwasaki[4], Naruhiko Adachi[5], Toshio Moriya[5], Masato Kawasaki[5], Katsumi Maenaka[1] ([1]*Fac. of Pharm. Sci., Hokkaido Univ.*, [2]*Operand OIL, AIST*, [3]*IPR, Osaka Univ.*, [4]*TARA, Univ. of Tsukuba*, [5]*SBRC, KEK*)

Insect lipids are stored in fat body and transferred to muscle or body surface by lipid transfer protein complex, lipophorin. Lipophorin carries phospholipids, cholesterol and diacylglycerol. Lipophorin is recycled after lipid transportation which is one of the unique features of lipophorin and different from human lipoproteins. To elucidate the molecular mechanism of lipophorin, we purified lipophorin from silkworm larvae and observed lipophorin particles by electron microscope. The lipophorin particle showed uniform spherical shape and the 2D class average result suggested that lipophorin was composed of two internal domains, one of which seems responsible for lipid binding.

2Pos009 HIV-2 糖タンパク質の構造機能解析
Structure and functional analysis of human immunodeficiency virus type-2 (HIV-2) envelope glycoprotein

Yuki Anraku[1], Shunsuke Kita[2], Hideo Hukuhara[2], Simon Davis[3], Atsushi Hurukawa[2], Thushan de Silva[3], James Robinson[4], Yuguang Zhao[3], Yvonne Jones[3], David Stuart[3], Juha Huiskonen[3], Sarah Rowland-Jones[3], Katsumi Maenaka[2] ([1]*Grad. Sch. Life Sci., Univ. Hokkaido*, [2]*Faculty of Pharm Sci., Univ. Hokkaido*, [3]*Univ. Oxford*, [4]*Univ. Tulane*)

Human immunodeficiency virus (HIV) is composed of two groups, HIV-1 and HIV-2. While HIV-1 is the main cause of HIV infections, HIV-2 patients are limited to West Africa. Therefore HIV-2 has not been well studied so far. To investigate the infectivity of HIV-2 in molecular level, we prepared HIV-2 envelope proteins (Env), and evaluated their oligomer states. The HIV-2 Env proteins were expressed with or without anti-HIV-2 antibody in HEK293T and purified from supernatants by affinity chromatography followed by gel filtration. Obtained HIV-2 Env proteins were analyzed using Blue Native-PAGE and electron microscope. I would like to discuss the result of molecular characteristics of HIV-2 Env proteins.

2Pos010 分子動力学シミュレーションを用いたαシヌクレインアミロイドの構造解析
Structural analysis of α-synuclein amyloids using molecular dynamics simulation

Hiroki Otaki, Yuzuru Taguchi, Noriyuki Nishida (*Grad. Sch. Biomedical Sci., Nagasaki Univ.*)

α-Synuclein (αSyn) amyloid is the main component of Lewy body, which is a hallmark of dementia with Lewy bodies (DLB), and also causes Parkinson's disease (PD) and multiple system atrophy (MSA). Recently, αSyn amyloids isolated from DLB and MSA have been reported to show distinct proteolytic fragment patterns, and to have transmissivity and strain diversity like prions. Structure of the αSyn amyloid has been analyzed using cryo-EM and suggested as in-register parallel β-sheet structure with a unique "Greek-key" conformation. In this work, we investigated conformational propensity of the wild type and mutant αSyn amyloids using molecular dynamics simulation, suggestive of distinct conformations, which may explain the mechanism of the diversity of amyloid.

2Pos011 翻訳後修飾によってシトルリン化したヒト S100A3 蛋白質の擬似体の探索
Exploring the posttranslational modification of human S100A3 protein using citrullination mimics

Kenji Ite[1,2], Kenji Kizawa[3], Kenichi Kitanishi[4], Masaki Unno[1,2] ([1]*Graduate School of Science and Engineering, Ibaraki University*, [2]*Frontier Research Center for Applied Atomic Sciences, Ibaraki University*, [3]*Kao Corporation*, [4]*Department of Chemistry, Faculty of Science, Tokyo University of Science*)

S100A3 protein consists of total 101 amino acids including four arginine residues per a molecule. Peptidylarginine deiminase type III (PAD3) converts a symmetric pair of Arg51, but not other arginines, to citrullines on dimeric S100A3 in the presence of Ca^{2+}. This specific citrullination of the S100A3 dimer causes formation of a homotetramer, and thereby cooperatively increases its affinity for Zn^{2+} and Ca^{2+}. To elucidate this mechanism, we aim to clarify structure of the Ca^{2+}/Zn^{2+}-bound citrullinated S100A3. However, the citrullinated form of S100A3 is difficult to prepare in large quantities. In this study, we prepared the Arg51 substituted mutants as mimics of S100A3 citrullinated by PAD3 and analyzed biophysical property of the prepared mutants.

2Pos012 機械学習を用いたタンパク質主鎖構造における 2 面角の再分類
Reclassification of dihedral angles in protein backbone structures using machine learning

Hiroto Murata, George Chikenji (*Dept. Appl. Phys., Nagoya Univ.*)

The Ramachandran plot is a fundamental tool in the analysis of protein structures and it has been often classified into several specific, well known regions, such as alpha-helix, beta-sheet, PPII region. These regions are, however, obtained by averaging amino acid types or by ignoring its neighbouring residue effects. Thus, there may be a possibility that some interesting or important regions have not been identified yet. Here, we used Gaussian Mixture Method to re-classify Ramachandran map depending on its amino acid type and neighbouring effects. As a result, we found that some interesting new regions that have been ignored so far.

2Pos013 糖化が LDL の物性に与える影響について
Effect of glycation on the physical properties of low density-lipoprotein

Seiji Takeda[1], Toshihiro Sakurai[1], Shu-Ping Hui[1], Hitoshi Chiba[2] ([1]*Faculty of Health Sciences, Hokkaido Univ.*, [2]*Faculty of Health Science, Sapporo University of Health Sciences*)

Oxidation of the low-density lipoproteins (LDLs) induce development of cardiovascular disease. We previously reported that elastic modulus of LDL is decreased after oxidation or phospholipase A2 treatment by using an atomic force microscopy (AFM). This study used AFM to investigate glycation effects on the elastic modulus. After LDL was obtained from human serum, it was modified in vitro using ribose or glucose. After glycation, glycated LDL was absorbed on a mica surface. The LDL topography and the elastic modulus were measured. Although the glycated LDL size was unchanged, the glycated LDL elastic modulus was lower. Moreover, binding affinity of the glycated LDL to mica surface was changed. This report presents discussion of the related mechanism.

2Pos014 酵母プリオン Sup35 の液-液相分離と線維化に対する共溶質の影響
Effects of co-solutes on liquid-liquid phase separation and fibrillization of yeast prion Sup35

Suguru Nishinami[1], Yumiko Ohhashi[2], Kentaro Shiraki[1] ([1]*Pure and Appl. Sci., Univ. Tsukuba*, [2]*Grad. Sch. Sci., Univ. Kobe*)

Prion disease is a serious neurodegenerative propagated via protein. Sup35 is a yeast prion protein with 3 domains: C-terminal domain has a translation termination factor, and N-terminal and middle (NM) domain has intrinsic disordered regions. NM domain matures to fibrils that lead to cell death, while it forms liquid-liquid phase separation in response to starvation to protect C domain for cell survival. However, a relation between droplets and fibrillization remained unclear. Here, we examined the effects of various types of co-solutes, such as chaotropic or kosmotropic ions, amino acids, osmolytes, and neutral or charged crowder, on various size assembly of Sup35. In this conference, we will discuss the effects of co-solutes on droplets and fibrils of Sup35.

2Pos015 局所クラスター間構造コンプライアンスに基づくタンパク質の形状に内在する変形伝播特性の解析
Analysis of the Deformation Transmission Properties in Protein Shapes based on the Structural Compliance between Localized Clusters

Keisuke Arikawa (*Fcl. Eng., Kanagawa Inst. of Tech.*)

The deformation of a localized structure in an object can cause the deformation of other localized structures. Herein, we propose a method for analyzing the deformation transmission (DT) properties in protein shapes using an elastic network model. In this method, we divide the model into clusters and evaluate the cluster deformation when forces are applied to the other cluster. The DT indices can be defined based on the structural compliance between a pair of clusters (one cluster can be assigned to the whole structure). Subsequently, we can identify the parts at which it is easy or difficult to transmit deformations by calculating the indices for all pairs of the clusters. Furthermore, the distributions of the indices are observed to characterize the protein shapes.

2Pos016 フェレドキシン構造とそのリバース構造の、PDB における発生頻度が異なるのはなぜか？
Why occurring frequencies of ferredoxin and its reverse fold in PDB are largely different?

Megumi Nakajima, George Chikenji (*Nagoya University Graduate School of Engineering Department of Applied Physics Sasai Laboratory*)

A superfold is defined as a protein fold which has been observed in three or more non-homologous proteins in the protein structure database. The most frequently observed superfold in the SCOP database is ferredoxin-fold, which consists of a four-stranded beta sheet and two alpha helices. Interestingly, the observed frequency of reverse ferredoxin, obtained by reversing only the N- to C-term chain direction of ferredoxin, is extremely small compared with that of ferredoxin, although these two folds are structurally quite similar to each other. The question arises is whether this difference comes from sampling bias of evolution or thermodynamic stability. Here we study the origin of the difference by the database analysis and physics based simulations.

2Pos017 Structural Characterization of β_2 Microglobulin Core Fragments in Amyloid Fibrils using Circular Dichroism Theory and Molecular Dynamics

Koichi Matsuo[1,3], Hirotsugu Hiramatsu[2], Robert W. Woody[3] ([1]*Hiroshima Synchrotron Radiation Center, Hiroshima University*, [2]*Department of Applied Chemistry, National Chiao Tung University*, [3]*Department of Biochemistry and Molecular Biology, Colorado State University*)

Circular dichroism (CD) spectroscopy combined with CD theory has great potential for characterizing the steric structures of backbone and aromatic side chains of proteins. CD calculations on 26 proteins from the crystal structures and the simulated structures using molecular dynamics (MD) revealed that the theoretical CD averaged over MD trajectories reproduced well the experimental CD compared to those of crystal structures. The CD spectrum of β_2-microglobulin core fragments in amyloid fibrils was measured from 300 to 178 nm at pH 5.0 and calculated under acid condition. The comparison between the experimental and theoretical spectra showed that the dihedral angles of disulfide bonds between fragments are crucial factors for determining the unique fibril conformations.

2Pos018 B 型肝炎ウイルス(HBV)への逆転写阻害薬剤分子のカプシド内部の自由エネルギー計算
Calculation of free energy of transfer of a reverse transcription inhibitor to the inside of Hepatitis B Virus (HBV) capsid

Ryo Urano[1], Kazushi Fujimoto[1], Yoshimichi Andoh[2], Noriyuki Yoshii[2], Wataru Shinoda[1], Susumu Okazaki[1] ([1]*Grad. Sch. Eng., Nagoya Univ.*, [2]*Center Comput. Sci. Grad. Sch. Sci., Nagoya Univ.*)

Hepatitis B virus (HBV) can cause serious symptom such as cirrhosis and liver cancer into hundreds of millions of infected people in the world every year. Although the virus belongs to a DNA virus, they replicate its DNA using reverse transcription in the lifecycle. An effective drug to this virus is nucleotide or nucleoside reverse transcription inhibitors (NRTIs) such as entecavir. On the other hand, current drugs have problems in the occurrence of drug mutant virus and side effect such as nephrotoxicity. Therefore, a more effective drug is required to cure patients. In this work, we perform the free energy simulation to evaluate the absorption of a drug in the liver cell into the pregenomic HBV capsid to investigate the capsid core at the molecular level.

2Pos019 アンチパラレルβシート中のジスルフィド結合が分泌タンパク質ホールディングに及ぼす影響
The effect of disulfide bonds in anti-parallel β-sheets on secreted protein folding

Hiromi Suzuki (*School of Agri., Meiji Univ.*)

We selected 21,312 protein chains from PDB and analyzed amino acid pairs located on the adjacent strands in β-sheets. Preference of non-hydrogen bond (nHB) Cys-Cys pairs in anti-parallel sheets was 11 times higher than expected value and was exceptionally higher than that of any other pairs including hydrogen bond Cys-Cys pairs. One of the reasons of such higher preference was that disulfide (SS) bonds formed between adjacent β-strands in anti-parallel sheets were formed only for nHB pairs with two exceptions. More than 70% of these SS bonds were formed in secreted and membrane proteins, suggesting that these SS bonds contributed to β-strand assembly in these proteins.

2Pos020 エンドセリン B 受容体の分子動力学シミュレーション
Molecular dynamics simulations of human endothelin B receptor

Koichi Abe[1], Yoshitaka Moriwaki[1], Kentaro Shimizu[1,2], Tohru Terada[2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

Endothelin (ET) is a 21-amino-acid peptide involved in vascular control and binds to ET receptors (ET_AR and ET_BR), class-A G-protein coupled receptors. Recently, crystal structures of the human ET_BR bound to ETs (ET-1 and ET-3), a partial agonist, and antagonists were determined. However, the crystal structures contain mutations and insertion for promoting crystallization. Here, we replaced the mutated residues of ET_BR with the original ones and performed microsecond molecular dynamics simulations to reveal the wild-type structures and their dynamics. We also performed modeling of the complex structure between TxET-1 and ET_BR, which has not been solved experimentally. We will discuss the difference in the structure between the agonist- and antagonist-bound states.

2Pos021 クライオ電子顕微鏡単粒子解析法を用いた KcsA の構造解析
Structural Analysis of KcsA by Cryo-EM Single Particle Analysis

Hiroko Takazaki[1], Hirofumi Shimizu[2], Takuo Yasunaga[1] ([1]*Grad. Sch. Comp. Sci. Syst. Eng., KIT*, [2]*Fac. Med. Sci., Univ. Fukui*)

KcsA is a potassium channel. It has open and closed conformations under acidic and neutral conditions, respectively. Both structures of full-length KcsA have been solved by X-ray crystallography. However, the structural changes between the two conformations are predicted to be more dynamic by diffracted X-ray tracking. In this study, we try to elucidate the structures of KcsA under open and closed conditions by cryo-electron microscopy (EM) and single particle analysis. To obtain the high-resolution structure, we investigated its monodisperse conditions in thin ice. As a result, in using amphipol, we implemented its monodispersity by gel filtration chromatography and observed two conformations. Now, we are analyzing KcsA structures with amphipol.

2Pos022 マスク付きセグメンテーション・フィット法による複数サブユニットの電顕マップへの局所重ね合わせ
Masked segmentation fitting of multiple atomic subunits into a local 3D EM density map

Takeshi Kawabata, Haruki Nakamura, Genji Kurisu (*Institute for Protein Research, Osaka University*)

Rigid-body fitting of subunits into a 3D density map is an important step for atomic modeling on electron microscopy. However, fitting multiple subunits on local regions of the map is still difficult, if some of the subunit models are missing. For the purpose, we propose a “masked segmentation & fitting” algorithm, using Gaussian mixture model. The algorithm iterates following two steps. The “segmentation” step assigns subunit labels on regions of the map. The “fitting” step superimposes each subunit only on the corresponding labeled region of the map. Repeating these steps efficiently fits multiple subunits on entire region of the map. For the local fitting, we introduced a masking region around each subunit atomic model for the segmentation step.

2Pos023* 糸状仮足の構造変化とアクチンフィラメントの分布の相関
Correlation between structural changes of filopodia and distribution of actin filaments

Miho Nakafukasako, Tomoya Higo, Yusuke V. Morimoto, Takuo Yasunaga (*Grad. Sch. Comp. Sci. Syst. Eng., KIT.*)

Cells perform cell movement using the filopodia. Our previous study has suggested that there are two types of structures of filopodia, i.e. "Round structure" with a rounded tip and “Sharp structure” with formed up by actin filament bundles to the tip. In this study, to elucidate the molecular mechanism of cell movement, filopodial actin filaments were structurally analyzed. We observed the dynamics and structure of actin filaments inside the filopodia in NG108-15 cells using fluorescence microscopy and cryo-electron microscopy. We evaluated the correlation between the filopodial structure and cell movement, including actin distribution and suggested how the manner of actin filaments inside should induce their formation.

2Pos024* Does the secondary site of neuraminidase play any significant role in drug resistance of influenza?

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In influenza treatment, drugs that inhibit the enzymatic activity of neuraminidase (NA) in influenza virus, such as oseltamivir and zanamivir, have been used. After treatment of influenza with these NA inhibitors, drug-resistant strains are known to appear at about 20%. A major outstanding issue in understanding the drug resistance mechanism of influenza virus is the role of a secondary site adjacent to the active site on NA. The secondary site attracts not only substrate but also inhibitors stronger than the active site, and thus may play a role in drug resistance of influenza; however, its molecular mechanism has been unclear. In this study, we intend to clarify the drug resistance mechanism of influenza virus, focusing on the role of the secondary binding site of NA.

2Pos025* 多次元 NMR 分光法を用いたヤエヤマサソリ由来殺虫性ペプチド毒素 LaIT2 の機能領域の溶液構造解析
Structural studies for the functional domains of insecticidal peptide toxin, LaIT2, with heteronuclear multidimensional NMR spectroscopy

Chiharu Tatsushiro[1], Maiki Tamura[2], Hironori Juichi[3], Masahiro Miyashita[3], Hisashi Miyagawa[3], Shinya Ohki[2], Hayato Morita[1,4] ([1]*Fac Sci., Josai Univ*, [2]*Grad. Sch. Mat Sci., JIAST*, [3]*Grad. Sch. Agr., Kyoto Univ*, [4]*Grad. Sch. Sci., Josai Univ*)

Liocheles australasiae is the one of two Japanese scorpions and mainly inhabits in the Yaeyama island. *L. australasiae* has the series of peptide like poison, those specifically acting for insects. Among these poisons, LaIT2(59AA) is not only insecticidal, but also has antibacterial activities. Until now, we have found that the N-terminal and C-terminal domains of LaIT2 has different physiological functions. In this study, we have constructed the *E.coli* overexpression systems for these two domains and prepared the ^{15}N labelled domains. Furthermore, we have measured the heteronuclear multidimensional NMR spectra and analyzed the relationships between functions and structures of these domains.

2Pos026* 分子動力学計算で探る p53 の C 末端部位の DNA 結合機構
DNA recognition mechanisms of the p53 C-terminal domain Investigated by MD simulation

Yuta Taira[1], Duy Tran[1], Jacob Swadling[2], Akio Kitao[1] ([1]*Tokyo Tech*, [2]*Univ.Tokyo*)

p53 is a tumor-suppresser protein that binds to DNA by the core binding domain and intrinsically disordered C-terminal domain. The latter domain non-specifically binds to DNA but the binding mechanisms are still unclear. In order to elucidate the mechanism of the DNA recognition by the p53 C-terminal domain, we prepared different initial structures of the domain distributed around a model DNA duplex, and simulated p53-DNA binding by tens of independent all-atom MD simulations. As a result, we observed p53 sliding along the DNA in atomic level and identified some preferred binding sites on the DNA structure. Also, we examined the relation between the p53 structure and DNA binding. We further investigated DNA binding affinities of amino acid residues in p53.

2Pos027* 天然変性タンパク質 Tau の溶液中における過渡的な凝集原繊維構造形成
Intrinsically disordered protein Tau tends to transiently form a part of the protofilament core structure in the soluble state

Ryosuke Kawasaki[1], Shin-ichi Tate[2] ([1]*Dept. MLS, Grad. Sch. Sci., Hiroshima Univ.*, [2]*Prog. MLS, Grad. Sch. Integr. Sci. for Life, Hiroshima Univ.*)

Intrinsically disordered proteins (IDPs) do not have well-defined three-dimensional structures. Through the transformation from soluble fraction to aggregation, IDPs play key roles in the development of neurodegenerative diseases. Microtubule binding protein Tau is one of the IDPs and plays major roles in microtubule (MT) stabilization in the neuron. In Alzheimer's disease brain, Tau detached from MT forms aggregation, leading loss of its function. The detailed mechanism to form insoluble aggregation is not understood. According to solution NMR, I could detect the transient folding like a part of the protofilament core structure of PHF in the soluble state. Thus I propose the aggregation mechanism through the transient structure of IDPs in solution.

2Pos028 Single-molecule FRET experiments for investigation of DNA single-strand damage recognition mechanism by PARP-1

Anna Sefer[1], Eleni Kallis[1], Tobias Eilert[1], Mara Guariento[1], Nadine Jakobi[1], David Neuhaus[2], Sebastian Eustermann[3], Jens Michaelis[1] ([1]*Ulm University, Institute of Biophysics, Albert-Einstein Allee 11, 89081 Ulm, Germany*, [2]*MRC Laboratory of Molecular Biology, Francis Crick Avenue, Cambridge CB2 0QH, UK*, [3]*Ludwig-Maximilians-University Munich, Gene Center and Department of Biochemistry, Feodor-Lynen-Strasse 25, 81377 Munich, Germany*)

Poly-(ADP-ribose)-polymerase-1 (PARP-1) is a key DNA damage signaling protein that detects DNA single strand breaks (SSBs). Upon PARP-1 binding to a SSB (nick or gap), the DNA adopts a highly kinked conformation. The fluorescent labels on the damaged DNA are positioned such that different degrees of DNA kinking lead to different Foerster Resonance Energy Transfer (FRET) efficiencies depending on different DNA conformations. Here we aim to investigate the mechanism of SSB recognition by PARP-1 and especially the associated conformatioanal changes of the DNA on the single molecule level via single molecule FRET experiments. Furthermore, we quantify the kinking angles of the DNA using a combination of computational approaches and the described smFRET experiments.

2Pos029 (2SGP-3) Determination of protonated states for native and mutant structures of HIV-1 protease with indinavir by free energy calculations

Masahiko Taguchi, Ryo Oyama, Masahiro Kaneso, Shigehiko Hayashi (*Kyoto University*)

HIV-1 protease is a target of drug development and various inhibitors binding to two aspartic acids in the catalytic site were developed. However, it has a serious drug-resistance. To resolve the drug-resistance problem, it is importnat to determine protonated states of the two aspartic acids. For one of typical inhibitors, indinavir, it is known that mutation (V82T/I84V) greatly affects its binding free energy. To clarify the structural difference between native and mutant proteases with indinavir, we performed QM/MM free energy calculations. Then we succeeded to determine protonated states for both structures precisely, and revealed a change of hydrated environment around catalytic site by the mutation.

2Pos030 Crucial role of conformational excitation in enzyme catalysis of Pin1

Toshifumi Mori[1,2], Shinji Saito[1,2] ([1]*IMS*, [2]*SOKENDAI*)

Conformational flexibility of protein is essential for enzymes. Yet, how this contributes to catalysis remains highly controversial. Pin1 is a member of the peptidyl-prolyl isomerase; NMR experiments find micro- to millisecond dynamics in Pin1, yet the role of this dynamics in cis-trans isomerizations is unclear. Here we show how Pin1's dynamics contribute to catalysis in molecular detail using molecular dynamics simulations. Conformational rearrangement of Pin1 about the ligand is found to be essential for stabilizing the transition state, yet isomerization transitions occur rapidly (~ps); protein's slow dynamics thus do not follow isomerization. Instead, the catalytic conformation of Pin1 is prepared a priori, i.e. as a conformational excitation out of equilibrium.

2Pos031 Weighted ensemble simulations of the cis-trans isomerization in Pin1 enzyme using the QM/MM method

Norifumi Yamamoto[1], Kei Moritsugu[2], Yasushige Yonezawa[3], Shin-ichi Tate[4], Hiroshi Fujisaki[5] ([1]*Chiba Tech*, [2]*Yokohama City Univ*, [3]*Kindai Univ*, [4]*Hiroshima Univ*, [5]*Nippon Med Sch*)

Proline cis-trans isomerization is known to play critical roles in protein folding, splicing, and cell signaling, etc. Pin1 is a highly conserved peptidyl-prolyl isomerase, which has been implicated in many diseases such as cancer and Alzheimer's. In this study, we try to clarify detailed catalytic mechanisms of Pin1 using molecular dynamics simulations. We perform the weighted ensemble (WE) simulations for the isomerization based on an empirical force field to extract some kinetic information such as the mean first passage time. To further refine the model, we proceed to employ a QM/MM hybrid model to examine the reliability of empirical force fields and discuss the further applications of WE simulations to enzymatic reactions.

2Pos032 Simulation Study on Atomistic and Physicochemical Properties of Amyloid beta 42

Ikuo Kurisaki, Shigenori Tanaka (*System Info., Grad. Schl., Kobe Univ.*)

Recently, oligomer formation of amyloid beta 42 (A-beta(42)) draws attention for its toxicity to neuronal cells. Meanwhile, because of broad-spectrum population of oligomer species, it is challenging to elucidate atomistic and physicochemical properties for oligomer with a specific size. We addressed this problem by employing all-atom molecular dynamics simulations for A-beta(42) nonamer. We observed positional twisting between protomers. Twisting of the protomers results in reduction of activation barrier of protomer-protomer dissociation. Lowering activation barrier could be due to weakened interaction between them. This observation indicates that protomer dissociation reaction proceeds via conformational change of A-beta(42) nonamer.

2Pos033 敵対的生成ネットワーク（GAN）を用いた新規主鎖構造のタンパク質デザイン
Protein design with novel main-chain structure using Generative Adversarial Networks

Takaaki Sato[1], Yoshitaka Moriwaki[1], Tohru Terada[2], Kentaro Shimizu[1,2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

Introducing non-structural information such as evolutionary information as distance restraints is very effective in the *de novo* protein structure prediction. Recently, it is demonstrated that the DCGAN can generate artificial pairwise distance matrices from the whole PDB structural data and the Cα configurations can be generated by solving convex optimization problems allowing a certain range of error. Here we implemented this method with the state-of-the-art GANs and generated full-atom models using Rosetta from the Cα configurations. We found that the Cα configurations generated by our implementation were more accurate than those generated by the original method. We are now planning to apply this method to *de novo* protein design.

2Pos034 テルペン環化酵素における基質と反応の選択性に関する計算科学的研究
Computational investigation of the substrate and reaction selectivity of terpene cyclases

Masanobu Arita[1], Keiichi Murai[1], Yoshitaka Moriwaki[1], Tohru Terada[2], Tomohisa Kuzuyama[1], Kentaro Shimizu[1,2] ([1]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [2]*III, Univ. Tokyo*)

Terpenoids are the most abundant and largest class of natural products. Type-I terpene cyclases convert linear precursors (geranyl diphosphate, farnesyl diphosphate, or geranylgeranyl diphosphate) into a large structural variety of (poly)cyclic terpene hydrocarbons or alcohols. However, the products and the substrates of bacterial terpene cyclases cannot be predicted from their amino acid sequences alone. Here, we have developed a method that combines molecular dynamics simulation and Rosetta docking to elucidate the mechanism of the specificity. With this method, we can predict important residues responsible for the recognition of the substrate and the stabilization of the intermediate states in the enzyme.

2Pos035 Pin1 由来のタンパク質分解酵素のミクロ化
Micronization of a protease derived from Pin1

Teikichi Ikura, Nobutoshi Ito (*Med. Res. Inst., Tokyo Med. Dent. Univ.*)

A peptidyl-prolyl isomerase, Pin1, functions as a switch in the cell cycle, a regulator for the Alzheimer's disease, and so on. We found that some mutations like C113A deactivated the isomerase activity but created a proteolytic activity. Since then, we have investigated the catalytic mechanism of the novel protease derived from Pin1. So far, we have elucidated that this protease recognizes a proline residue and the major catalytic site was composed of typical triad seen in some serine proteases. In addition, molecular dynamics simulation of the mutant suggests that WW domain has no contribution to the proteolytic catalysis. In the present study, we removed the WW domain, and introduced additional mutations to improve the catalytic activity and stabilize the protein.

2Pos036 緑色硫黄細菌 ferredoxin-NADP$^+$酸化還元酵素と基質間の酸化還元反応の特異性
Unique kinetic behavior in the redox reaction catalyzed by ferredoxin-NADP$^+$ oxidoreductase from green sulfur bacteria

Daisuke Seo (*Grd. Sch. Nat. Sci. Tec., Kanazawa Univ.*)

Ferredoxin-NAD(P)$^+$ oxidoreductase (FNR) is a flavoprotein catalyzing the redox reaction between the small iron sulfur protein ferredoxin (Fd) and NAD(P)H reversibly. FNR from *Chlorobaculum tepidum* (CtFNR) is a unique homo-dimeric flavoprotein homologous to the bacterial NADPH-thioredoxin reductase (TrxR). Pre-steady state kinetic analyses of the CtFNR reduction by reduced Fd in the presence of NADP$^+$ revealed that the transition of redox state of CtFNR substantially affected the observed rate for the NADP$^+$ reduction. Obtained transient spectra suggested the difference in the conformation of FAD for reduced CtFNR from other TrxR-type FNRs. The catalytic mechanism of CtFNR will be discussed by comparing with other TrxR-type FNRs.

2Pos037 タンパク質キナーゼへの ATP 競合阻害剤結合の自由エネルギー解析
Free energy analysis of ATP competitive inhibitor-protein kinase bindings

Suyong Re, Hiraku Oshima, Yuji Sugita (*RIKEN Center for Biosystems Dynamics Research*)

Recent drug compound design focuses not only a ligand's binding affinity to a target protein but also its residence time (kinetics). The latter requires knowledge on the binding pathway, which is hardly obtainable from traditional X-ray crystal structure analysis. Here, we apply two-dimensional replica-exchange (gREST/REUS) binding simulations to ATP competitive inhibitor bindings to Src protein kinase, an important drug target. The simulations reveal free-energy landscapes of the bindings and show an important contribution of early stage encounter complex formations to the bindings. Further details on the binding mechanism and implication for drug compound design would be discussed.

2Pos038 フォトンファクトリーにおける生体高分子溶液試料の小角 X 線散乱に関する発展
Progress of Biological Small-Angle X-ray Scattering at the Photon Factory

Kento Yonezawa, Masatsuyo Takahashi, Keiko Yatabe, Yasuko Nagatani, **Nobutaka Shimizu** (*KEK, IMSS, PF*)

Biological small-angle X-ray scattering (BioSAXS) is versatile and efficient technique to obtain information of both low-resolution structure and properties on biological molecules and their complexes in solution. Thus, it is actively utilized as one of various methods in hybrid approach today. SAXS beamlines at the Photon Factory are compatible with the latest BioSAXS technologies such as SEC-SAXS, and we have developed and released an automatic analysis software for SEC-SAXS data, *Serial Analyzer*[1]. We also have supported new users for measurement and analysis of BioSAXS based on BINDS[2]. We will introduce about recent progress and works in this presentation.

1. *AIP Conf. Proc.* **2054**, 060082 (2019). doi: 10.1063/1.5084713
2. https://www.binds.jp/

2Pos039 抗体修飾ナノニードルを用いた生細胞における中間径フィラメントの可動性解析
Mobility analysis of intermediate filament in a living cell using antibody-functionalized nanoneedle and AFM

Ayana Yamagishi[1,2,5], Moe Susaki[1,2], Mei Mizusawa[1,2], Akira Nagasaki[1], Saku Kijima[3], Q.P. Taro Uyeda[1,4], **Chikashi Nakamura**[1,2,5] ([1]*Biomed. Res. Inst., AIST*, [2]*Dept. Biotechnol. & Life Sci., Grad. Sch. Eng., TUAT*, [3]*Bioproc. Res. Inst., AIST*, [4]*Dept. Phys., Sch. Adv. Sci. Eng., Waseda Univ.*, [5]*PhotoBio-OIL, AIST-Osaka Univ.*)

Nestin, an intermediate filament protein, has been implicated in cancer metastasis. Here, we established a nestin-knockout (KO) cell line to evaluate effects of nestin disruption on a highly metastatic mouse breast cancer cell. Because the KO cells were significantly stiffer than parental cells, nestin presumably increases the invasiveness by softening the cell body. We speculate that vimentin when copolymerizes with nestin is inhibited to bind actin filaments due to the large size of the nestin tail domain, and reduction in the binding sites increases the flexibility of the cytoskeletal structure. To test this hypothesis, we also evaluated mobility of the vimentin filament in a living cell measured by using antibody-functionalized nanoneedle and AFM.

2Pos040 Investigation on the relationship between cytotoxicity and amorphous oligomers

Punitha Velmurugan, Jannatul Aklima, Yoshihiro Ohta, Yutaka Kuroda (*Tokyo university of Agriculture and Technology*)

The cytotoxicity of the amyloid oligomers is actively explored, but the impact of amorphous oligomers remains unaddressed. Reliable and reproducible production of amorphous oligomers with given size and biophysical properties remain a challenging task. Solubility controlling peptide tag (SCP-tag, a short 3-7 residue peptides) is emerging as a way to control amorphous oligomer formation with relatively high reliability. Here, we report cytotoxicity of amorphous oligomers of D3ED3 (Domain 3 of DENV3 envelope protein), produced using a four Ile-tag (C4I-tag), against HeLa cells. At the meeting, we will report the size of the oligomers in cell medium after incubation for 0 and 24 h as measured by Dynamic Light Scattering, as well as cell viability as monitored by MTT assay.

2Pos041 In vitro ATPase-based screening of circadian clock mutants of KaiC in cyanobacterial circadian clock system

Dongyan Ouyang[1], Atsushi Mukaiyama[1,2], Yoshihiko Furuike[1,2], Kumiko Miwa[3], Takao Kondo[3], Shuji Akiyama[1,2] ([1]*Inst. Mol. Sci.*, [2]*The Grad. Univ. for Adv. Studies*, [3]*Grad. Sch. of Sci., Nagoya Univ*)

KaiC is a core protein of the cyanobacterial circadian clock. A low and temperature-insensitive ATPase activity of KaiC is considered as one of the factors governing the temperature-compensated period length of the clock system. To identify its structural unit responsible for the clock functions, we developed an in vitro ATPase-based screening system. So far, we have evaluated 86 of 519 single-alanine substitutions in KaiC and found diverse mutants carrying activated or inactivated ATPases. for validation purposes, we picked up three mutants from our library and characterized their clock functions both in vitro and in vivo. we would like to discuss the effect of mutations in relation to the KaiC structure.

2Pos042 構造蛋白質である HIV-1p17 と p24 の動的と静的構造の解析
Dynamic and rigid structures of HIV-1 p17 and p24 proteins

Chiaki Nishimura (*Fac. Pharm. Sci., Teikyo Heisei Univ.*)

p17 matrix and p24 capsid proteins are sequentially synthesized at the N-terminal part in Gag Polyprotein in the infected cell by HIV-1. One of the activation processes of two proteins is to be cleaved into two proteins by the HIV-1 protease. The NMR studies revealed the possibly responsible structure, where p17 has the flexible structure at the C-terminal regions including helix 5 and C-terminal tail. On the other hand, the N-terminal beta-structures were required in p24 N-terminal domain. Furthermore, the isomer formation at Proline 90 is essential for the cyclosphilin-A binding of p24. It was concluded that minor form of cis-conformation is important to recognize the cyclophilin-A. Thus, the structure-function relationship of p17 and p24 proteins will be discussed.

2Pos043 アミロイドベータタンパク質を分解する人工ペプチドの設計
Designing artificial peptides that hydrolyze amyloid beta protein

Yoshihiro Iida, Atsuo Tamura (*Kobe Univ, Grad Sch Sci*)

Amyloid beta protein is known to be the cause of Alzheimer's disease. It has been found that this protein is cytotoxic when it forms an oligomer. Therefore, we aimed to improve Alzheimer's disease by artificially designing a peptide that hydrolyzes the oligomeric form of amyloid beta protein. First, we created a peptide that is random coiled when isolated, but takes on an alpha-helix structure when bound to amyloid beta protein to form the conformation of the catalytic triad. This peptide was found to selectively hydrolyze the amyloid beta protein and to inhibit its membrane disruption capability. It is concluded that the designed peptide can be regarded as a protease which is capable of hydrolyzing amyloid beta protein.

2Pos044 4 つのイントロン位置は、アスパラギン酸アミノ転移酵素立体構造上で平面を形成する。
Four intron positions form a plane in the tertiary structure of aspartate aminotransferase

Michiko Nosaka (*N.I.T., Sasebo College*)

We have identified intron-planes about different proteins. Although we have discussed about the possible reason of this plane, the meaning or function of this phenomenon is not clear at this time. Here, we report the characteristics of the planes of aspartate aminotransferase and discuss about the hypothesis to explain. 1, The probability of forming a plane is large enough for this medium size enzyme and two planes are formed. 2, Almost all atoms of substrate analog with Co-enzyme of the protein is included in one of the plane, whose probability is very low (average of 20 cycles is about 0.4%). 3, While in the other plane of this enzyme, it contained most atoms of the ligating residue for the Co-enzyme in free condition from its substrate.

2Pos045 抗微生物ペプチド Cryptdin-4 の多量体の脂質分子存在下における分子シミュレーションによる観察
Antimicrobial peptide Cryptdin-4 oligomers interacting with lipids observed by molecular dynamics simulations

Takao Yoda (*Nagahama Institute of Bio-Science and Technology*)

Cryptdin-4 (Crp4) is an antimicrobial peptide that is known for its high bactericidal activity. Crp4 not only permeabilizes negatively-charged membranes but also translocates across them. An experimental study suggested that oligomerization of Crp4 takes place on its translocation. Since it is monomer in solution, oligomers are transient. Therefore, we considered that molecular dynamics simulation is a powerful tool to study its oligomerization and translocation processes. Here we performed 320-ns lipid self-organization simulations 53 times independently with two to seven Crp4 molecules included in the simulated systems, with different simulation conditions. We analyzed oligomer conformations found in the simulation trajectories.

2Pos046 ヘム ABC インポーター BhuUV-T の構造変化の自由エネルギー解析
Free energy analysis for the conformational changes of a heme ABC importer BhuUV-T

Koichi Tamura[1], Yuji Sugita[1,2,3] ([1]*RIKEN R-CCS*, [2]*RIKEN TMS*, [3]*RIKEN BDR*)

Heme importer BhuUV-T is a member of the type-II ATP-binding cassette (ABC) transporters which transport heme across the membrane bilayer of a bacteria. Recently, crystal structures of BhuUV-T in the nucleotide-free inward-facing form have been solved. Based on the structures and biochemical experiments, a molecular mechanism for the heme transport cycle was proposed. In this study, a computational modeling approach is adopted to predict the occluded intermediate with bound nucleotides. The predicted model is further validated by the free energy analysis based on the string method. The computational results support the proposed transport cycle of BhuUV-T.

2Pos047* トレハロースによるミオグロビンの構造安定化および酸性条件下でのアミロイド形成からの回復作用
Stabilization of the Myoglobin Structure and Restoration from the Amyloid Formation under Acidic Conditions by Trehalose

Satoshi Ajito[1], Mitsuhiro Hirai[1], Nobutaka Shimizu[2], Noriyuki Igarashi[2] ([1]*Grad. Sch. Sci. Tec., Gunma Univ.*, [2]*KEK*)

Sugars are well-known to prevent protein denaturation. In particular, trehalose has received attention because of its association with the adaptation of organisms to extreme environments. Recently, we reported that sugars preserve the protein hydration shell and stabilize the protein structure against chemical and thermal denaturation. This study focuses on the restoration of acid-denatured amyloid formation of myoglobin by trehalose. By using the wide-angle X-ray scattering method, we found that myoglobin at an initial process of amyloidogenic reaction (helix-to-sheet transition followed by oligomerization) was substantially restored to its native structure by trehalose. In addition, the results of the effect of sugars on cold denaturation of protein will be presented.

2Pos048* 異なる緩衝剤中での抗体の安定性と構造変化の関係
Relation between stability and structure of an antibody in different buffers

Hiroaki Oyama[1], Kanta Enomoto[1], Tetsuo Torisu[1], Susumu Uchiyama[1,2] ([1]*Grad. Sch. Eng, Osaka Univ*, [2]*ExCELLS*)

It has been recently recognized that antibody aggregates may cause adverse effects such as immunogenicity as well as decrease in drag efficacy. Optimizations of antibody drug formulation to minimize aggregation are required. Thus mechanistic understanding of buffer contribution to the stability could pave the way for the formulation development from empirical screening-based approach to rational selection. In this study, acetate buffer (AB) and citrate buffer (CB) at pH 5.0 which are often used in antibody drug formulation were examined. By employing classical but robust physico-chemical approaches, this study clarified the difference in the antibody stability in two buffers at same pH but different chemical formula from micro- and macroscopic viewpoints.

2Pos049* ショウジョウバエ Argonaute2 の N 末端領域はアミロイド繊維を形成する
N-terminal region of Drosophila Argonaute2 can form amyloid fibrils

Haruka Narita, Makoto F. Kuwabara, Tomotaka Komori, Ryo Murakami, Tomohiro Shima, Mikiko C. Siomi, Soutaro Uemura (*Department of Biological Sciences, Graduate School of Science, The University of Tokyo*)

Argonaute plays a crucial role in RNA silencing. Drosophila Argonaute2 (DmAgo2) contains an unusual N-terminus insertion (N-ter) with unclear function. Because its amino acids sequence has a characteristic feature of prion-like domain (PrLD), here we experimentally tested and confirmed that purified N-ter aggregates into amyloid fibrils even in the presence of SDS, indicating that N-ter is a PrLD. Further, this amyloid fibril was disaggregated by heat shock proteins (HSPs), which is known to regulate DmAgo2's RNA silencing activity. In this meeting, we will discuss how HSP-related aggregation/disaggregation of N-ter regulates RNA silencing by DmAgo2.

2Pos050* Dynamics of the helix-coil transition of alanine-based polypeptides detected by nanosecond region fluorescence correlation spectroscopy

Supawich Kamonprasertsuk[1,2], Hiroyuki Oikawa[1,2], Satoshi Takahashi[1,2] ([1]*IMRAM.Tohoku Univ.*, [2]*Grad.Sch.Sci.,Tohoku Univ.*)

Helix-coil transition is an elementary process involved in protein folding. The time constant of the transition mostly deduced from temperature jump experiments, however, varies from several nanoseconds to a few microseconds. To examine the dynamics by a different method, we utilized fluorescence correlation spectroscopy and investigated the alanine-based peptide labeled by donor and acceptor dyes. The method is based on the analysis of the time intervals of two photons and allows the detection of nanosecond dynamics in equilibrium conditions. We detected ~70 and ~500 ns dynamics in the absence and presence of trifluoroethanol, respectively, that likely correspond to the helix-coil transitions. We will discuss the comparison of the current and previous time constants.

2Pos051 MD simulation along with MSM analysis reconstructs LPA_6 binding pathway

Rieko Hirota[1], Ryuichiro Ishitani[1], Mizuki Takemoto[1,2], Osamu Nureki[1] ([1]*Dept. of Biosci., Grad Sch. of Sci., Univ. of Tokyo*, [2]*Present address: Preferred Networks, Inc.*)

Lysophosphatidic acid (LPA) receptors are G-protein coupled receptors (GPCRs) which are related to such as cell differentiation or migration. Recently, our group determined the X-ray crystal structure of the LPA receptors, LPA_6 which mainly contributes to hair formation. This structure has a groove which laterally opens towards the lipid membrane, suggesting that LPA might approach from the membrane and bind to the biding site laterally via this groove. This new binding model has not been examined enough yet. Here, we succeeded to reconstruct the full pathway of LPA_6-ligand binding by molecular dynamics (MD) simulation along with Markov state model analysis. This result implied that K185 and K188 assist the LPA hydrophilic head entry into the binding pocket.

2Pos052 加熱による中性 pH での β_2 ミクログロブリンのアミロイド線維形成
Heating-induced amyloid formation of β_2-microglobulin at neutral pH

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Amyloid fibrils associated with various amyloidoses are one of the misfolding aggregates formed by denatured proteins. β_2-Microglobulin (β2m) forms amyloid fibrils and causes dialysis-related amyloidosis (DRA) in patients receiving long-term hemodialysis. Although DRA develops under physiological conditions, amyloid formation of β2m in vitro has been difficult at neutral pH because of the resistant native structure. Here, we found that β2m efficiently forms amyloid fibrils even at neutral pH by heating under agitation. We showed the importance of supersaturation for the amyloid formation and constructed the temperature- and NaCl concentration-dependent conformational phase diagrams. Our results suggest a critical role of heating in the onset of DRA.

2Pos053 Functional Sensitivity and Mutational Robustness of Proteins

Qianyuan Tang, Tetsuhiro Hatakeyama, Kunihiko Kaneko (*Grad. Sch. Art. & Sci., Univ. Tokyo*)

Native proteins can have both high susceptibility in the functional dynamics and high robustness in the evolution. For native proteins, local perturbations can be felt persistently by all residues of the protein. Meanwhile, such dynamics are highly robust to minor differences in protein structure or detailed interactions at the atomic level, showing that such kind of sensitive response is robust in the evolution. To establish the physical link between functional sensitivity and mutational robustness, we set up a general framework to analyze the response of proteins under internal and external perturbations. By entropy maximization approach, we further demonstrate that power-law distribution will emerge in the normal mode distribution of native proteins.

2Pos054 多次元仮想座標とカップルした分子動力学法を用いた mSin3 複合体の立体構造探索
Conformational sampling of an mSin3 complex using multidimensional virtual-system coupled canonical MD

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NRSF/REST is a transcription repressor regulating neural genes. N-terminal-side repressor domain binds to a corepressor mSin3 and recruits a histone deacetylase (HDAC). A chemical compound that inhibits the binding of flexible PAH1 domain of mSin3 and NRSF/REST can be a drug candidate for neural diseases. Therefore, the compound is an important target for the drug discovery research. We performed a generalized-ensemble Molecular Dynamics simulation of mSin3 PAH1 with or without a ligand and analyzed the free-energy landscapes to assess their interactions and realistic binding mechanism. Multidimensional virtual-system coupled canonical MD (mD-VcMD) was used to enhance conformational sampling to explore a high-dimensional reaction-coordinate space.

2Pos055 アデニル酸キナーゼの構造転移の改良カメレオンモデルによる研究
Conformational transition of adenylate kinase studied with the improved chameleon model

Ryota Mori, Masaki Sasai, Tomoki P. Terada (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

Proteins exhibit conformational transition related to their biological functions. To improve our understanding of the conformational transition, we have previously proposed the chameleon model, which is a coarse-grained model representing the interdependence of local structural changes during conformational transitions (Terada et al., J. Phys. Chem. 2013). With some modification, we have applied this model to adenylate kinase, a model protein of large-scale domain movement. Langevin dynamics simulation yielded free energy landscape consistent with two-state conformational transition with domain movement similar to the experimental observation. We also discuss the structure of transition state ensemble in terms of local structural changes and local unfolding.

2Pos056 Spectroscopic analysis of protein crowded environments using the charge-transfer fluorescence probe ANS

Chikashi Ota[1], **Kazufumi Takano**[2] ([1]*Ritsumeikan Univ.*, [2]*Kyoto Pref. Univ.*)

In this study, the analytical method to evaluate the crowding effect has been suggested by using a charge-transfer fluorescence probe, ANS. By employing the unique property of ANS attaching to charged residues on the surface of lysozyme, the crowding effect was focused, while the case was compared as a reference, in which ANS is confined in hydrophobic pockets of BSA. Consequently, the surface specific changes of fluorescence spectra were readily observed under the crowded environment, whereas the fluorescence spectra of ANS in protein inside did not change. This result suggests the fluorescence spectra of ANS binding to protein surface have the capability to estimate the crowding effect of proteins.

2Pos057 (2SHA-6) ダイナミン GTP アーゼはアクチン線維の束化と分散を機械的に制御する
(2SHA-6) Dynamin GTPase mechanically regulates bundling and unbundling of actin filaments

Kohji Takei[1], La The Mon[1], Tadashi Abe[1], Tetsuya Takeda[1], Ikuko Fujiwara[2], Akihiro Narita[3] ([1]*Grad.Sch. Med.Dent. Pharm.Sci., Okayama Univ.*, [2]*Dept. Biol.Facul.Sci., Osaka City Univ.*, [3]*Struct. Biol. Res. Ctr and Divi. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

Actin cytoskeleton is dynamically remodeled in variety of cellular events. We previously found that dynamin bundles F-actin (Yamada et al. J Neurosci. 2013; Int. J. Oncol. 2016). However, the molecular mechanisms are poorly understood. Here we show that dynamin mechanically bundles F-actin by polymerizing into helix around several actin filaments. Dynamin GTPase activity was drastically enhanced by the presence of F-actin. Addition of GTP to F-actin bundles formed by dynamin lead to rapid depolymerization of dynamin and unbundling of actin filaments. Furthermore, we found that ionic strength affects both dynamin polymerization and actin bundle formation. Thus, bundling and unbundling of F-actin is strongly linked to dynamin's self-polymerization and GTP hydrolysis.

2Pos058 FRET リアルタイム活性測定系を用いた大腸菌 S2P ファミリー膜内切断プロテアーゼ RseP の kinetics 解析
Kinetic analysis of the proteolytic reaction catalyzed by S2P family intramembrane protease RseP using a FRET-based real-time assay system

Yohei Hizukuri, Yoshinori Akiyama (*Inst. Front. Life Med. Sci., Kyoto Univ.*)

RseP, an *Escherichia coli* member of S2P family intramembrane proteases, has a critical role in activation of extracytoplasmic stress response through cleavage of anti-σ membrane protein RseA. Although a detailed kinetic analysis provides an important basis to understand the functions of the proteolytic enzymes, such study has not been conducted for the S2P family proteases. Here we report construction of a FRET-based real-time activity assay system for RseP using a synthetic peptide substrate. We designed a peptide having FRET pair probes at the end regions of the RseA transmembrane sequence. The synthesized peptide was degraded by purified RseP. Real-time monitoring of the FRET signals enabled kinetic analysis of the RseP-catalyzed substrate proteolysis.

2Pos059 Recognition mechanism of proteins which bind to versatile amino acid sequences

Katsumi Omagari (*Dept. of Virology, Medical School, Nagoya City University*)

DNA-damage-binding protein 1 (DDB1) has several cellular substrates of which binding sites have diverse amino acid sequences and bind to viral proteins also. To reveal the mechanism in which those substrates bind to DDB1 despite sequence diversity, the binding of DDB1 to the substrates in living cell was examined with protein-protein visualization system, Fluoppi. The Fluoppi visualizes Protein-protein interactions as fluorescent puncta (FP) in living cells. The viral proteins and substrates in association with DDB1 forms clear FP in living cell. Those binding kinetics indicated that the binding affinities of the viral proteins for DDB1 has much higher than the one of the substrates in living cells. Viral proteins might optimize the binding affinity to DDB1.

2Pos060 呼吸鎖超複合体形成タンパク質によって制御される Cyt *c* のパートナータンパク質認識機構
The mechanism of cytochrome *c*-redox partner proteins recognition regulated by the respiratory supercomplex factor protein

Wataru Sato[1], Koichiro Ishimori[2], Peter Brzezinski[1] ([1]*Stockholm Univ. Fac. of Nat. Sci.*, [2]*Hokkaido Univ. Fac. of Sci.*)

In respiratory chain, cytochrome c (Cyt c) shuttles an electron from cytochrome bc1 complex (Cyt bc1) to cytochrome c oxidase (CcO), but the physical mechanism how the Cyt c selectively recognizes these redox partner proteins in cell remains unclear. Here, we explored the mechanism regulating the Cyt bc1-CcO electron transfer (ET) cycle via Cyt c in terms of interactions of Cyt c with Cyt bc1-CcO supercomplex factor protein, Rcf1. Various kinetic analyses found that the Cyt c oxidation in the Cyt c-CcO complex and the re-binding of Cyt c to Cyt bc1 were promoted in the presence of Rcf1, suggesting the contribution of Rcf1 to a unidirectional transfer of Cyt c from CcO to Cyt bc1 through the interactions with oxidized Cyt c, leading to an efficient Cyt bc1-CcO ET cycle.

2Pos061 QM/MM 法によるニワトリ卵白リゾチームの糖加水分解反応シミュレーション
QM/MM Study on Hydrolysis of Polysaccharides in Hen Egg-White Lysozyme

Takuya Uto[1], Yoshiki Mitani[2], Toshifumi Yui[2] ([1]*Organization for Promotion of Tenure Track, University of Miyazaki*, [2]*Faculty of Engineering, University of Miyazaki*)

Hen egg-white lysozyme (HEWL) is well known as endoglycosidase which degrades the polysaccharide of bacterial cell walls. The glycosidic linkage between carbohydrate residues located at sub-sites +1 and -1, is cleaved through the cooperative reaction of Glu35 and Asp52. Most recently, we have successfully reproduced the whole reaction scheme of HEWL-catalyzed hydrolysis by adopting the QM/MM simulation. It was observed that the covalent intermediate was formed between the anomeric carbon of N-acetylglucosamine (NAG) and Asp52 through the cleavage of glycosidic linkage by proton donation from Glu35. Subsequently, water dissociation induced the pyranose ring conversion from chair form to boat form accompanied with breaking covalent bond between NAG and Asp52.

2Pos062 幾何学に基づく新しいタンパク質構造解析プログラム
A new program to analyze protein structures based on the geometric context

Anri Terabayashi[1], Momoka Nakamura[1], Kyosuke Sakata[1], Takuya Miyakawa[2], Masaru Tanokura[2], Tohru Terada[3], Masaki Kojima[1] ([1]*Sch. Life Sci., Tokyo Univ. Pharm. Life Sci.*, [2]*Grad. Sch. Agric. Life Sci. Univ. Tokyo*, [3]*III, Univ. Tokyo*)

We have developed a series of program package VOLTES (Virtual Optimization of Local TErtiary Structures), to analyze protein structures based on not the physicochemical but the geometric (especially topological) context. In VOLTES1 program, all the protein structures are expressed as torsion angles of rotatable bonds, and the torsion-angle values are arranged into 'trees' having the same topologies as the proteins' structures. In addition, each torsion angle is discretized into 6 classes to reduce the number of conformations to be considered. We applied VOLTES1 to two proteins with different folds, calaxin and neoculin. The results showed the program was very effective to detect the crucial rotatable bonds responsible for their global conformational changes.

2Pos063 細胞質中 RAF 分子の構造およびダイマー化状態を捉える 1 分子計測
Conformational and dimeric states of cytosolic RAF detected by single-molecule measurements

Kenji Okamoto[1], Kayo Hibino[2], Yasushi Sako[1] ([1]*RIKEN CPR*, [2]*NIG*)

Function of proteins are intimately related to molecular states, such as conformation, chemical modification or oligomerization. Such states are often complexly regulated by interactions with other molecules in live cells. We have successfully applied single-molecule Förster resonance energy transfer (smFRET) measurement using alternative laser excitation (ALEX) technique to cytosolic C-RAF proteins and acquired various quantities, such as FRET and fluorescence intensity, from single RAFs. The results indicated the existence of at least three structural states and transitions among them. It was also implied that homodimers of C-RAF were formed in live cells and they play important roles in RAF's function, such as response to stimulation by epidermal growth factor.

2Pos064 信号変化の順序決定の自動化およびタンパク質 HD 交換ダイナミクスの振動バンドへの応用
Automatic determination of the sequential order of signal changes and its application to vibrational bands of protein H-D exchange process

Daisuke Miyata[1], Takakazu Nakabayashi[1], Shinichi Morita[2] ([1]*Graduate School of Pharmaceutical Sciences, Tohoku University*, [2]*Graduate School of Science, Tohoku University*)

Automated method for determining the sequential order of arbitral signal changes was developed in generalized two-dimensional (2D) correlation spectroscopy. Conventionally, for spectral changes, the synchronous 2D correlation spectrum and the asynchronous 2D correlation spectrum were calculated, which gave information on the sequential order between two bands. For multiple bands (here, ca. 30 bands), the sequential order was determined manually to cause complicated and time-consuming situations. We developed the automated method, which was applied to infrared spectra of H-D exchange process of lysozyme. This method automatically distinguished the rate constants of the side chains and those of secondary structures of the main chain.

2Pos065 水素高感度解析を実現するタンパク質中性子回折実験の進歩
Progress of the protein neutron diffractometry to realize hydrogen high sensitivity analysis

Ichiro Tanaka (*Graduate School of Science and Engineering, Ibaraki University*)

Hydrogen atoms play a crucial role in biological systems, including enzymatic reactions, but the presence cannot be identified in the X-ray crystal structures. Next-generation neutron sources (e.g., J-PARC) enable us to carry out neutron diffraction analysis of proteins, which provides us a lot of useful information of hydrogen atoms. It will be presented that new techniques for neutron measurement such as high pressure freezing and dynamic nuclear polarization of protein have been developed, and that there is a plan of new neutron facility to gain more S/N ratio so that the sample crystal volume can be much small to find protonation/deprotonation in the near future.

2Pos066 Flexible Fitting of Biomolecular Structures to Atomic-Force-Microscopy Images via Biased Molecular Simulations

Toru Niina, Sotaro Fuchigami, Shoji Takada (*Grad. Sch. Sci. Univ. Kyoto*)

Atomic force microscope (AFM) is a prominent and useful tool to observe large-scale structural dynamics of biomolecules at the single molecule resolution without loss of their functionalities. However, the data obtained by AFM are limited to the surface shape of the molecules. Here, we developed the method to fit a three-dimensional structure model into an AFM image. First, we modeled the AFM measuring process by using a combination of smooth functions. Next, we developed a potential energy function that is capable to achieve flexible fitting into AFM images. As a validation, we tested our method using synthetic AFM images. We then applied the method to an experimental result. We confirmed that our method is capable to find the correct orientation and conformation.

2Pos067 Deep convolutional neural networks for identifying cryo-EM grid holes suitable for particle collection

Yuichi Yokoyama[1], Tohru Terada[3], Kentaro Shimizu[2,3], Kazutoshi Tani[4] ([1]*GSII, Univ. Tokyo*, [2]*Grad. Sch. Agr. Life Sci., Univ. Tokyo*, [3]*III, Univ. Tokyo*, [4]*Grad. Sch. Med., Univ. Mie*)

Cryo-electron microscopy(cryo-EM) is surging in popularity as a method for protein structure determination. High-resolution structure determination by cryo-EM single-particle analysis requires a large number of particle images in vitreous ice of suitable thickness suspended across grid holes. We have developed a pipeline for automatically identifying suitable holes using convolutional neural networks. This pipeline is composed of two models: a one-stage object detection model to detect all grid holes from low-magnification images and an object recognition model to classify the holes according to labeled training images. Our pipeline is much faster than manual selection and closely matches human performance, enabling rapid protein structure determination.

2Pos068 アクチン線維に張力を発生させると線維の長軸まわりのねじれは減少する
Mechanical stress declined the amplitude of the torsional fluctuations of single actin filaments

Kaoru Okura, Takumi Fukuda, Hitoshi Tatsumi (*Department of Applied Bioscience, Kanazawa Inst. of Technol., Ishikawa, Japan*)

It has been proposed that actin filaments sense tension and act as a mechanosensor. F-actin was polymerized from G-Actin labeled Alexa-555. A magnetic bead was attached to an actin filament, and was pulled toward an electrical magnet to generate tension in the filament. We directly measured the torsional fluctuations of single actin filaments along its long axis with single molecule fluorescence polarization microscopy. When tension was generated, the amplitude of torsional fluctuations was decreased.

2Pos069 高速 AFM 画像のドリフト除去法の応用
Application of drift elimination method for high-speed AFM images

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High-speed atomic force microscopy (HS-AFM) enables real-time imaging of dynamics of biomolecules under near physiological conditions [M. Shibata et al., Nat. Commun. 2017.]. However, HS-AFM images often have a position shift (drift) unrelated to the function of proteins. This is because weakly adsorbs to the AFM substrate so as not to inhibit the function of a target protein. In this study, we applied the motion correction software in RELION, which makes great achievements in single-particle analysis of Cryo-EM. As a specific target, we used HS-AFM images of Ca2+/calmodulin-dependent protein kinase II (CaMKII), which plays an important role in learning and memory. In the poster, we will show structural changes of CaMKII due to the binding of Ca2+/calmodulin.

2Pos070* ポリプロリンロッドの挿入によるドメインスワッピング二量体のデザイン
Poly-proline rod insertion for the design of domain-swapped dimer

Shota Shiga[1], Masaru Yamanaka[2], Wataru Hujiwara[1], Shun Hirota[2], Shuichiro Goda[3], Koki Makabe[1] ([1]*Graduate School of Science and Engineering, Yamagata University*, [2]*Division of Materials Science, Nara Institute of Science and Technology*, [3]*Graduate School of Engineering, Nagasaki University*)

Domain-swapping is one of the mechanisms for protein oligomerization via interchanging its structural element. Here, we report a simple design strategy for creating an artificial domain-swapped dimer from monomeric protein by inserting a poly-proline rod at a deleted loop. The crystal structures revealed that the designed dimers formed a long extended shape. Surprisingly, the extended shape was retained in solution confirmed by the SAXS experiment, indicating the rigidity of the poly-proline rod insertion. The design strategy will expand the design possibility of protein oligomer. We also found that the crystals of the domain-swapped dimers have porous tube packing with high solvent content and we will discuss the designability of the pore in the crystal.

2Pos071* c-Myb-KIX 相互作用を阻害するペプチドの計算機によるデザイン
Computational design of a peptide inhibitor targeting c-Myb-KIX interaction

Shunji Suetaka[1], Yoshiki Oka[1], Tomoko Kunihara[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

The transcriptional activator c-Myb interacts with the KIX domain of CBP and regulates hematopoiesis. However, its aberrant expression causes leukemia. Here, we rationally designed peptide inhibitors of the c-Myb-KIX interaction by introducing mutations into the transactivation domain of c-Myb (c-Myb32). Because c-Myb32 is intrinsically disordered and binds KIX by the conformation selection mechanism, we designed the c-Myb32 mutants with their helices stabilized using theoretical helicity predictions, to increase the affinity to KIX. As a result, we succeeded in designing the peptide that binds KIX ~3-fold more tightly than the wild type. We are currently working on improving the peptide inhibitors using the Rosetta software.

2Pos072* 超安定タンパク質構造のゼロからの合理設計とその融合による、GPCR 構造の合理的安定化
Tailor-made design of superstable proteins from scratch for rational stabilization of GPCR

Masaya Mitsumoto[1,2], Nanao Suzuki[3], Ryosuke Nakano[3], Takahiro Kosugi[1,2,4], Takeshi Murata[3], Nobuyasu Koga[1,2,4] ([1]*ExCELLS, NINS*, [2]*SOKENDAI*, [3]*Fac. of Sci., Chiba Univ.*, [4]*IMS, NINS*)

G-protein coupled receptors (GPCRs) are one of the most important membrane proteins associated with physiological functions. Since GPCRs are innately unstable, increasing the stability is required for mass-production, solving structures, obtaining antibodies etc. In addition, state-controllability is required for solving structures in the aimed states. Here, we computationally designed all-alpha proteins consisting of 6-helices from scratch with the NC terminal helices to be seamlessly fused to GPCR structures in the active and inactive states. The designs highly expressed in E. coli are found to be extremely heat-stable in the circular dichroism, and the chimeric GPCR with them shows increased detergent resistance and heat-stability in the clear-native PAGE.

2Pos073* 理論的飽和変異解析によるジヒドロ葉酸還元酵素の高活性化
Improving activity of dihydrofolate reductase by theoretical saturation mutagenesis

Kazuhisa Ohara[1], Yoshiki Oka[1], Yuuki Hayashi[1], Munehito Arai[1,2] ([1]*Dept. Life Sci., Univ. Tokyo*, [2]*Dept. Phys., Univ. Tokyo*)

Theoretically improving activities of enzymes is expected to be extremely useful in industry. Dihydrofolate reductase (DHFR) has been used as a model enzyme in experimental and theoretical studies on the correlation between dynamics and function. In this study, we aimed to rationally enhance the activity of DHFR based on theoretical predictions. Using the protein design software "Rosetta", we performed theoretical saturation mutagenesis for all residues of DHFR and searched for the mutants that may accelerate the rate-limiting step in the catalytic cycle. We succeeded in designing the DHFR mutant that has 2.5-fold higher activity than the wild type. Thus, our design procedure can be a general method for improving activities of enzymes.

2Pos074* Engineering of genome editing protein Cas9 that slides along DNA faster and might enable efficient target search

Trishit Banerjee[1,2], Dwiky Rendra Graha Subekti[1,3], Hiroto Takahashi[1], Satoshi Takahashi[1], Kiyoto Kamagata[1] ([1]*IMRAM, Tohoku Uni.*, [2]*Fac. of Sci., Tohoku Uni.*, [3]*Grad. Sch. of Sci., Tohoku Uni.*)

Genome editing protein Cas9 has been used in various biological studies; however, Cas9 has two defects: low efficiency and inaccuracy. Our earlier experiment based on single molecule fluorescence microscopy revealed that catalytically inactive Cas9 (dCas9), slides along DNA to much lesser extent than other DNA binding proteins. In this study, we tried to engineer dCas9 to slide along DNA faster by adding a sliding-promoting peptide. We analyzed the engineered dCas9 using single molecule fluorescence microscopy and DNA-aligned array, DNA garden. The engineered dCas9 showed enhancement of diffusion constant by three times relative to dCas9. This strategy may help in reducing target-DNA search time by Cas9 and might lead to precise and efficient gene editing in the future.

2Pos075 (2SGP-8) 天然変性タンパク質 p53 を標的としたペプチドの人工設計ー液液相分離の制御ー
(2SGP-8) Rational design of peptide targeting intrinsically disordered protein p53—regulation of function and phase-phase separation—

Kiyoto Kamagata[1], Eriko Mano[1], Yuji Itoh[1], Saori Kanbayashi[1], Masaya Honda[1], Ryo Kitahara[2], Tomoshi Kameda[3] ([1]*IMRAM, Tohoku Univ.*, [2]*Coll. Pharmacy Sci., Ritsumeikan Univ.*, [3]*AIRC, AIST*)

Intrinsically disordered regions (IDRs) of proteins are involved in many diseases. However, the flexible IDRs hinders the use of 3D structure-based design methods. Here, we developed a rational design method to obtain a peptide that can bind an IDR using only sequence information. We applied the method to the disordered C-terminal domain of the tumor suppressor p53. The designed peptide showed the suppression of binding to DNA and sliding along DNA by p53 based on NMR, MD, and single molecule experiments. Also, we found that this peptide suppressed liquid liquid phase separation of p53 by blocking the interaction of the IDRs. Sequence-based design may be useful in designing peptides that target IDRs for therapeutic purposes.

2Pos076 膜内・膜外領域の改変によるサーモフィリックロドプシンの熱安定化
Further Thermo-Stabilization of Thermophilic Rhodopsin through Engineering in Intramembrane and Extramembrane Regions

Tomoki Akiyama[1], Naoki Kunishima[2], Masako Hirose[3], Sayaka Nemoto[4], Stoshi Yasuda[4,5,6], Yuki Sudo[7], Takeshi Murata[4,6] ([1]*Grad. Sch. Sci. & Eng., Univ. Chiba*, [2]*RIKEN RSC-Rigaku Collaboration Center*, [3]*Malvern Panalytical division of Spectris Co., Ltd*, [4]*Grad. Sch. Sci., Univ. Chiba*, [5]*Inst. Advanced Energy, Univ. Kyoto*, [6]*Molecular Chirality Research Center, Chiba University*, [7]*Grad. Sch. Med. Dent. Pharm. Sci., Univ. Okayama*)

Thermophilic rhodopsin (TR) is a light-driven proton pump with exceptional high thermostability. the hyperthermostable TR that functions at extremely high temperatures over 100 ℃ will expand its applicability as a thermostable biosensor in biotechnology. Here, we create the further thermostabilized TR by two different theories. As a result, 5 stabilized mutations were obtained in mutations designed by these methods. We determined the Tm of these stabilized mutations and WT by Differential scanning calorimetry(DSC), the Tm of all stabilized mutations was 2~4 ℃ higher than that of WT. We will try further stabilization of TR.

2Pos077 麹菌菌体外放出系の高機能化
Improvement of exocytotic secretion system of *Aspergillus oryzae*

Mone Kogure, Kensuke Nakajima, Yoshinori Tsuji, Yusuke Matsuda (*Dept.Biosci., Grad.Sch.Sci.Tech., Kwansei Gakuin Univ.*)

A. oryzae possesses an active lytic enzyme secretion system, and the system is applied for the heterologous protein productions. This study aimed at a development of systems for excretion of functional peptides and highly glycosylated proteins using *A. oryzae*. Gene sequences encoding Kex2-like protease cleavage site followed by *Oncorhynchus keta* (salmon) protamin or *Momordica charantia* (goya) lectin were linked to the 3' end of the intrinsic α-amylase gene and the construct was transformed into *A. oryzae*. Transformants were screened by nitrate auxotrophy, and genomic PCR. As a result of western blot analysis, accumulation of heterologous proteins within the cells were observed but yet with little excretion capacity.

2Pos078 細胞特異的ゲノム編集を目的とした核酸をデリバリーする新規一本鎖抗体 scFv の作製
Preparation of a novel single chain variable fragment (scFv) which delivers nucleic acid for cell-specific genome editing

Haruka Nasu[1], Yuji Sato[1], Takashi Tsukamoto[2,3], Takashi Kikukawa[2,3], Makoto Demura[2,3], Tomoyasu Aizawa[2,3] ([1]*Grad. Sch. Life Sci. Hokkaido Univ.*, [2]*Fac. Adv. Life Sci. Hokkaido Univ.*, [3]*GSS, GI-CoRE*)

Single chain variable fragment (scFv) is a molecule connecting two variable regions of light and heavy chains joined by short peptide linker. Because the scFv has lower molecular weight than antibody, it as a recombinant protein with functional sequence added can be prepared using Escherichia coli (E. coli). In this study, We developed a novel recombinant protein based on scFv to use of it for cell specific delivery of nuclease gene, one of important issues in genome editing technology. The recombinant protein consisting of scFv recognizing cell receptor and MPGH capable of condensing DNA was prepared using E. coli. As a result of investigating culture and purification conditions, a high purity recombinant protein could be obtained.

2Pos079 生体反応場におけるシトクロムｃ内多核ヘムの空間配置・酸化還元状態の変化
Cellular environment modulates geometry and redox state of deca-heme cofactors in bacterial surface cytochromes

Yoshihide Tokunou[1,2,3], Shingo Hattori[4], Thomas Clarke[5], Liang Shi[6], Kazuyuki Ishii[4], Akihiro Okamoto[2] ([1]*Faculty of Life. Environ. Sci., Uni. Tsukuba*, [2]*NIMS*, [3]*Research Fellow of JSPS*, [4]*Inst. Indust. Sci., Univ. Tokyo*, [5]*Centre for Molecular and Structural Biochemistry, Univ. East Anglia*, [6]*Dept. of Biological Sciences, China Univ. of Geosciences*)

Electron transport (ET) reactions proceeding in membrane proteins sustain biological energy acquisition. Biological ET reactions are driven under dynamic cellular environment in the presence of crowding biomolecules and continuous electron flow. However, analyses of ET reactions are limited to those with purified proteins under equilibrium condition. In this study, we obtained circular dichroism spectra of a decaheme cytochrome at outer-membrane (OMC) of intact Shewanella oneidensis MR-1, indicating that OMC changes its heme geometry and redox state specifically in intact cells. These cell-specific differences may alter ET kinetics and the redox potential of redox cofactors. In the presentation, we will discuss about ET kinetics data of MR-1 mutant strains.

2Pos080 NO-binding and protonation process in the catalytic reaction of the bacterial NO reductase as established by time-resolved spectroscopy

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NO reductase (NOR) is a membrane protein that reduces NO to N_2O at the active site, composed of heme and non-heme irons. The NO reduction mechanism has received great attention; however, it is yet to be defined because this reaction proceeds very fast. In this study, we tried tracking the catalytic reaction of NOR using time-resolved (TR) spectroscopy. Studying on not only native NOR but also its mutant, in which the proton transfer is suppressed, we could understand the timing of NO binding and proton transfer during the reaction. These findings allow us to propose a modified cis-heme mechanism. Currently, we are trying to characterize the coordination structure and electronic state of NO which bound to active site in reaction intermediates.

2Pos081 Anodized gold surface enables mediator-free bioelectrocatalysis of redox enzymes

Yasuhiro Mie, Yoshiaki Yasutake, Mashiki Ikegami, Tomohiro Tamura (*Bioproduction Res. Inst., AIST*)

Nanoporous gold (NPG) structure is attractive material in an electron transfer-based biophysical study owing to its large surface area, catalytic activity, high conductivity, and easiness for surface modification. We previously reported nanostructured dent surface is effective to drive human cyrochrome P450 (CYP). In the present study, NPG were demonstrated to be more efficient for mediator-free electron transfer of heme proteins including CYP and NAD^+-dependent enzymes. The NPG used was prepared by simply anodizing the commercially-available gold electrode, indicating the usefulness of the present system to utilize the enzyme function in various applications and understand the molecular basis in the function as well.

2Pos082* 膜蛋白質複合体の構造ダイナミクスへコレステロールが及ぼす影響に関する分子動力学的解析
Influences of cholesterol on structural dynamics of membrane protein complexes studied by molecular dynamics simulations

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Protein-protein interactions of membrane proteins play an important role in biological systems. Whereas it is known that their dynamics are closely related to lipids compositions of membrane, principles of this relationship are not fully clarified. This study aimed to clarify how lipid affects the structural dynamics of protein-protein interactions using molecular dynamics (MD). We simulated various complexes consisting of single- or two-helix Ala-Leu based model peptides and performed totally 20 μs MD simulations. As a result, cholesterol suppressed fluctuation of membrane proteins and affected the tilt angle of the protein to the membrane. In addition, effects of GXXXG motifs were also been discussed.

2Pos083* Observation of a β-Hairpin Peptide in α-Hemolysin Nanopore

Misa Yamaji, Ryuji Kawano (*Tokyo University of Agriculture and Technology*)

We report the electrical observation of single β-hairpin peptide in α-hemolysin (αHL) nanopore at the single molecule level. We have previously reported that hairpin-DNA (hpDNA) was inserted into the nanospace of αHL pore and observed its movement by the blocking current signals. We have also attempted to apply this system to sense nanospace that can estimate the environment of confined space by observing the movement of hpDNA as the nanopore probe. In this study, we try to establish the peptide probe instead of hpDNA. The β-hairpin peptide we synthesis and called as SV28 can remain the αHL nanopore and show the specific current signature. Furthermore, we analyzed the signal to analyze the insert-direction and blocking states of SV28 in the nanospace.

2Pos084* 表面プラズモン共鳴法を用いた膜タンパク質に特異的な脂質の同定: 脂質の生理機能解明を目指して
Identification of membrane proteins-specific lipids using surface plasmon resonance analysis: For elucidating the physiology of lipids

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Although it has been demonstrated that membrane proteins (MPs) required membrane lipids for MP functional and structural integrity, details on how MP-lipid interactions affect MP nature have yet to be fully understood. Previously, we developed the surface plasmon resonance (SPR)-based method for quantitatively evaluating the interactions, which enabled us to identify MP-specific lipids that affect MP activity. We applied this method to KcsA potassium channel from Streptomyces lividans to characterize the lipid-KcsA interactions, and eventually we proposed the mechanism of KcsA activation by lipids. Our presentation will show the detailed mechanism of KcsA activation, related to the interaction with lipid and the physical property of membrane.

2Pos085 クライオ電子顕微鏡による組換リアノジン受容体の高分解能構造
High-resolution cryo-EM structures of recombinant ryanodine receptors

Takuya Kobayashi[2], Akihisa Tsutsumi[3], Kei Saito[4], Takashi Sakurai[2], Masahide Kikkawa[3], Takashi Murayama[2], **Haruo Ogawa**[1] ([1]*IQB, The Univ. Tokyo*, [2]*Juntendo Univ. Grad. Sch. Med.*, [3]*Grad. Sch. Med., Univ. Tokyo*, [4]*Grad. Sch. Arts Sci., Univ. Tokyo*)

Ryanodine receptor (RyR) is a Ca2+ release channel in the sarcoplasmic reticulum of skeletal and cardiac muscles and plays a key role in excitation-contraction coupling. Here, we present high-resolution cryo-EM structures of recombinant RyRs. Recombinant RyRs were heterologously expressed in HEK293 cells, purified and observed under cryo-EM. The structure of RyR2 was determined at an average resolution of 3.3 Å, and, especially, local resolution around the transmembrane region was 2.9 Å, thus we could model atomic structure with no ambiguity. Because of the use of recombinant RyR, our approach has great potential to resolve the mechanisms by which many disease-associated mutations cause disease and may ultimately lead to the treatment of such diseases.

2Pos086 Exploring Structural Dynamics of Bacterial ABC Transporter MsbA by High Speed AFM

Kien X. Ngo, Holger Flechsig, Noriyuki Kodera, Toshio Ando (*WPI Nano Life Science Institute, Kanazawa University*)

MsbA, an ATP-binding cassette (ABC) transporter, exports lipid A across the cytoplasmic membrane of Gram-negative bacteria. MsbA also exports drugs, conferring multidrug resistance. The cryo-EM structures of MsbA bound with different nucleotides have suggested that large conformational changes possibly take place in the transmembrane domains (TMDs) coupled to the ATPase reaction in the ATP-binding sites formed by the two nucleotide binding domains (NBDs). In this study, we have succeeded in real-time imaging of structural dynamics of individual MsbA molecules during ATPase cycles by high speed AFM. The observed dynamic conformational changes in TMDs accompanying ATP hydrolysis in NBDs unveil a very important operational mechanism in bacterial ABC transporters.

2Pos087 Corynebacterial “Force-From-Lipids” mechanosensation for glutamate production

Yoshitaka Nakayama[1], Ken-ichi Hashimoto[2,3], Hisashi Kawasaki[2,3], Boris Martinac[1,4] ([1]*Victor Chang Cardiac Research Institute*, [2]*Biotech. Res. Cen., Univ. Tokyo*, [3]*Collab. Res. Ins. Inno. Microbiol., Univ. Tokyo*, [4]*University of New South Wales*)

A long-elusive puzzle of Corynebacterium glutamicum L-glutamate production has been solved since the mechanosensitive channel MscCG was identified as the major glutamate efflux system. The patch-clamp recording from C. glutamicum giant spheroplasts revealed the existence of three types of mechanosensitive channels including MscCG. All mechanosensitive channels are activated by increased membrane tension, indicating that the gating by mechanical force follows the “Force-From-Lipids” principle. Given that membrane lipids are altered significantly during the glutamate fermentation process, this suggests that the MscCG mechanosensitive channels also respond to changes in force transmission according to this principle.

2Pos088 The assembly of the trimeric autotransporter transmembrane domain into BamA-embedded nanodiscs

Eriko Aoki, Kazuo Fujiwara, Masamichi Ikeguchi (*Dept. of Bioinfo., Soka Univ.*)

Haemophilus Influenzae adhesin (Hia) is a trimeric autotransporter, which is the outer membrane protein (OMP) of gram-negative bacteria and mediates the adhesion to host cells. Although OMPs are thought to be inserted into the outer membrane by a β-barrel assembly machinery (BAM) complex, the insertion mechanism by BAM complex has not been fully elucidated. In this study, we investigated the assembly of Hia transmembrane domain (HiaTD) using empty and BamA-embedded nanodiscs. No assembly of HiaTD was observed when the empty nanodisc was used. In contrast, HiaTD was assembled into the trimer when BamA-embedded nanodisc was used. The result shows that BamA facilitates the assembly of HiaTD in vitro.

2Pos089 CRISPR Cas3 と Cse1 複合体の分子動力学シミュレーション
Molecular dynamics simulations of CRISPR Cas3 and Cse1 complex

Tomohiro Yamaguchi, Yui Taketomo, Naoyuki Miyashita (*BOST KINDAI Univ.*)

CRISPR system is usually worked as adaptive Immune system in prokaryotic cells. A portion of a genome from a virus is acquired into CRISPR system in the cell. Then, CRISPR uses it to find the genome from virus, and nick the DNA. CRISPR system has several types, and Type I CRISPR system is the most prevalent system. Type I CRISPR consists of several Cas proteins as modules. Cas3, is one of the modules, has the activity of nuclease and helicase. Cas3 is bind to the Cse1 in Type I CRISPR. Though the dynamics of Cas3 is one of the key proteins for the cleaving of DNA, the detail dynamics of the DNA-nicking is not know well. In this study, we performed the Molecular Dynamics simulations of Cas3 and Cse1 complex, to understand the DNA-nicking and helicase dynamics.

2Pos090 Identification of proteins that interact with nucleosomes by Quantitative Proteomics

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There are many different types of nucleosome, whose substitutes for the core histones (H3, H4, H2A, H2B) are altered to variants or partially removed, providing specific structural and functional features. We expect that these diverse nucleosomes present unique functions and are implicated in specific gene expression regulation. We have reported the structures of various nucleosomes such as H2A.Bbd nucleosome and overlapping dinucleosome and so on. However, the correlation between the structures and functional properties of these nucleosomes still remains unclear. In order to identify the interactome of the various nucleosomes, we conducted quantitative proteomics by LC-MS/MS. Novel proteins that interact specifically with the nucleosomes will be discussed.

2Pos091 転写因子の振る舞いとクロマチンのゆらぎの関係性を 1 分子計測によって解析する
Single molecular dynamics of transcription factors are controlled by diffusion movement of chromatin

Kazuko Okamoto[1], Yasushi Okada[1], Kuniya Abe[2], Tomonobu M Watanabe[1] ([1]*RIKEN BDR*, [2]*RIKEN BRC*)

Transcription factors are thought to facilitate the efficiency of target genes' transcription, and then regulate cellular states. However, the dynamics of transcription factors are poorly understood. To clarify the dynamics of transcription factors in living nuclei, we observed transcription factors Nanog and Oct4 at single molecular level in living nuclei of mouse ES cells along differentiation, and estimated the dwell time at target sites and the geometry of target sites. Our results suggested that both the dwell time and geometry change by the cellular state. Furthermore, the change in dwell time might be regulated by chromatin compaction. Our observation will link the behavior of transcription factors and chromatin dynamics.

2Pos092 DNA 二重鎖切断に応答した DNA トポイソメラーゼ 2B の核内挙動
Dynamic behavior of DNA topoisomerase 2B in response to DNA double-strand breaks

Ken ichi Yano, Keiko Morotomi-Yano (*IPPS, Kumamoto Univ.*)

DNA topoisomerase 2 (Top2) is crucial for resolving topological problems of DNA and plays important roles in various cellular processes. Because the involvement of Top2 in DNA repair remains unclear, we investigated the dynamic behavior of human Top2B in response to DNA double-strand breaks (DSBs), which is the most harmful DNA damage. Live cell imaging coupled with site-directed DSB induction by laser microirradiation demonstrated rapid recruitment of EGFP-tagged Top2B to the DSB site. FRAP analysis revealed that Top2B is highly mobile in the nucleus, and the Top2B catalytic inhibitors reduced the Top2B mobility and thereby prevented Top2B recruitment to DSBs. Collectively, our results highlight a novel aspect of Top2B functions in DSB repair.

2Pos093* 人工細胞デバイス内に封入した長鎖 DNA 1 分子からの遺伝子発現
Gene expression from a single large DNA encapsulated in artificial cell device

Yuto Ochiai[1], Hiroshi Ueno[1], Masayuki Su'etsugu[2], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*Dept. Life Sci., Col. Sci., Rikkyo Univ.*)

Reconstitution of autonomous artificial cells requires encapsulation of a large DNA and components for gene expression and replication into cell-sized reactors. In this work, we developed the artificial cell device system, in which cell-free protein synthesis from a 200 kbp DNA was implemented. We succeeded in imaging 200 kbp DNA encapsulated in a cell-sized reactor (φ=5 μm) and confirming *in vitro* gene expression simultaneously. Quantitative analysis revealed that most of 200 kbp DNA may keep its size, but, in some reactors, Venus was synthesized from fragmented DNA which couldn't be detected. Interestingly, Venus was synthesized from 200 kbp DNA with comparable yields and synthesis rate of those from 3 kbp DNA fragment.

2Pos094* 生体ナノポアフィルタを用いた DNA の一分子分離
Separation of a single molecule DNA using biological nanopore filter

Asuka Tada, Ryuji Kawano (*Department of Biotechnology and Life Science, Tokyo University of Agriculture and Technology*)

Single molecule experiments are essential to investigate the properties of individual molecules. In general, the single molecule observation and manipulation technique have been used for single molecule analysis. Although molecular separation or extraction at the single molecule level are also required, they are still under challenge. Here, we try to establish a biological nanopore filter for separation of target DNA at the single molecule level. In this system, we applied nanopore measurements, which can count DNA by the current blocking events. Additionally, we designed the capture DNA, which hybridizes with non-target DNA to pass through only target DNA. This system will be a useful tool for quantitative analysis of DNA at the single molecule level.

2Pos095* DETECTION OF DNA-POINT-MUTATION USING BIOLOGICAL NANOPORE

Ping Liu, Keisuke Shimizu, Ryuji Kawano (*Tokyo University of Agriculture and Technology*)

This paper describes the detection of DNA-point-mutation by using biological nanopore at the single molecule level. Identification of a point mutation in polynucleotides is essential for a diagnosis. Here we attempt to utilize nanopore technology for detecting a point mutation. In our strategy, a mutated DNA hybridizes to a probe DNA, and then the double-stranded DNA (dsDNA) is captured in the nanopore, and pass through pore with unzipping strands electrophoretically. In this study, we observed a difference of the unzipping duration, which caused by different DNA hybridization energy between WT and mutated DNA, as information to identify point mutation of dsDNA. Next, we will optimize the experimental condition to recognize the position of point mutation.

2Pos096 DNA を利用した人工分子モーターの実現を目指して
Towards the realization of artificial molecular motor using DNA molecules

Kohei Arai, Yuki Tsushima, Shoichi Toyabe (*Appl. Phys., Tohoku Univ.*)

The molecular motors are composed of functional domains such as the ATPase moiety. Developments in experimental techniques have been characterizing those domains in detail. However, it remains to be elucidated how the domains work together coordinately to achieve the robust and efficient function of the motor. In this project, we aimed to demonstrate an artificial molecular motor using DNA strands to understand how the elasticity of the motor body affects the motor's coordinated operation. DNA strands enable us to design not only the structure but also the interaction between molecules. For the first proof of concept, we designed an artificial motor composed of DNA and a micron-sized colloidal particle.

2Pos097 染色体脱凝縮シミュレーションによるヒト間期核組織化の理解
Organization of interphase human nucleus via simulated chromosome decondensation

Shin Fujishiro, Masaki Sasai (*Dept. Appl. Phys., Nagoya Univ.*)

Mammalian chromosomes spatially compartmentalize into active and repressed chromatin regions in interphase nucleus. While recent studies suggested phase separation of chromatin regions as the mechanism, it is still unclear how the nuclear scale compartments are formed. We have developed a computational polymer model of chromosomes where chromatin segments are coarse-grained into repulsive soft cores having different densities. Langevin simulations showed that non-equilibrium decondensation process of mitotic chromosomes predicted the compartmentalization and other organizations in human nucleus. Our results suggest the chromatin density-based mechanism of chromosome compartmentalization through decondensation.

2Pos098 クロマチン構造と RNA 輸送経路が協調して形成される過程
Cooperative formation of RNA transporting pathway and chromatin structure

Nozomu Imai, Shin Fujishiro, Masaki Sasai (*Dept. Appl. Phys., Nagoya Univ.*)

mRNA and other RNAs involved in translation are transported from the interior of nucleus through nuclear pore to cytoplasm, but it is still unclear how the pathway of RNA transport is formed in the nucleus. In this study, we simulate a chromatin system to analyze how the RNA transporting pathway is formed. The system is composed of heterochromatin and euchromatin regions near the nuclear membrane, RNA, and nuclear pores, and their movement is simulated with the Langevin equation. We investigated the regions where RNA passes through to reach the nuclear pore; the regions with the frequent residence of RNA depend on the parameters of diffusion and RNA-chromatin interactions, suggesting that RNA transporting pathway largely depends on structure and features of chromatin.

2Pos099 染色体のナノスケール 3 次元内部構造を可視化するための液中 3 次元原子間力顕微鏡（3D-AFM）の開発
Development of 3D-AFM to visualize nanometer-scale three-dimensional structures of chromosomes in liquid

Keisuke Miyazawa[1], Makiko Meguro-Horike[2], Shin-ichi Horike[2], Takashi Sumikama[3], Taku Higasayama[1], Masayuki Harada[1], Takeshi Fukuma[1,3] ([1]*Kanazawa Univ.*, [2]*Division of Functional Genomics, Advanced Science Research Center, Kanazawa Univ.*, [3]*Nano Life Science Institute (WPI-NanoLSI), Kanazawa Univ.*)

Chromosomes are three-dimensional (3D) structures containing DNA, histones and proteins with gene function of an organism. To observe 3D structures of chromosomes, electron microscopy has been extensively used. However, there are some technical limitations for precise observations of chromosomes such as a metal coating and vacuum environment. In this study, we develop 3D atomic force microscopy (3D-AFM) to visualize nanometer-scale three-dimensional structures of chromosomes in liquid. We fabricated thin (< 30 nm) and long (>500 nm) cylinder tip to insert a tip in a chromosome. The obtained 3D-AFM images show local contrasts correlating to part of inside structures of a chromosome. We are aiming to visualize whole 3D structures of chromosomes in liquid by 3D-AFM.

2Pos100 染色体の 3D-AFM 像と実像の関係の理論的解明
A theoretical study on a relationship between 3D atomic force macroscopy image and structure of chromosomes

Takashi Sumikama[1], Keisuke Miyazawa[4], Adam Foster[1,2,3], Takeshi Fukuma[1,4] ([1]*Nano Life Sicence Institute (WPI-NanoLSI), Kanazawa University*, [2]*Department of Applied Physics, Aalto University*, [3]*Graduate School Materials Science in Mainz*, [4]*Division of Electrical Engineering and Computer Science, Kanazawa University*)

Chromosomes are the molecules coding genes; however, their structurers at the nanometer to sub-micrometer scales are not fully understood yet. We are planning to measure them by using the three-dimensional atomic force microscopy (3D-AFM), but a relationship between 3D-AFM images and actual fiber structure of chromosome should be clarified to do so. Here, we made fractal globule structures of chromosomes in the interphase by a simple polymer simulation and developed a method to compute 3D-AFM images of them. It was found that some parts of fiber structure are really resolved in the 3D-AFM images but the others are hard to see, so we are applying a machine learning technique to fully recognize their detailed structures from the 3D-AFM images.

2Pos101 FRET study on sequence-dependent unwrapping of nucleosomal DNA

Tomoko Sunami, Di Luo, Hidetoshi Kono (*MMS, iQLS, QST*)

The first nucleosomes in the downstream of the transcription starting sites, +1 nucleosomes, have been found to play critical roles in genome organization. We have previously proposed that consecutive AA/TT dimer sequences in the entry side of +1 nucleosomes tend to unwrap more easily than those without AA/TT (Luo et al, NAR, 2018). To reveal sequence-dependent unwrapping in more detail, we conducted FRET measurement of reconstituted nucleosomes with sequences containing AA/TT repeat or containing the other dimer repeats. Our time-correlated single photon counting analysis showed that (1) the open state population is the most abundant in the nucleosomes with AT-rich sequences and (2) the open state is the most widely opened on the nucleosomes with long AA/TT repeat.

2Pos102 DNA メチル化に依存したヌクレオソームのスライディング動態の解析
Computational Analysis of the Nucleosome Sliding Dynamics Depending on DNA Methylation

Takeru Kameda[1,2], Miho Suzuki[3], Akinori Awazu[1,4], Yuichi Togashi[1,2,4] ([1]*Department of Mathematical and Life Sciences, Hiroshima University*, [2]*RIKEN Center for Biosystems Dynamics Research.*, [3]*Graduate School of Medicine, Nagoya University*, [4]*Graduate School of Integrated Sciences for Life, Hiroshima University.*)

Currently, our research group focus on methylation dependent nucleosome dynamics. In our previous experiment using Ciona intestinals, we obtained following result; (i) Gene body of half of the gene were methylated, and nucleosome sliding tend to decrease; (ii) Almost all of the methylated genes were housekeeping genes; These facts suggest that DNA methylation contributes gene expression regulation through the change of nucleosome dynamics. We discuss the details of the change of nucleosome dynamics from our current experimental results, performed by fully atomic molecular dynamics simulation.

2Pos103* 微小空間内における 1 分子からの長鎖 DNA 複製
Large DNA amplification from single molecule in micro-sized droplet

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Genetic information of single microorganisms is necessary for revealing the functions of the microorganisms. For the analysis, many DNA copies should be prepared. However, most species are hard to be cultivated artificially, DNA extracted from a microorganism should be amplified *in vitro*. Despite the development of various methods for DNA amplification, conventional methods have limitations of DNA length. Here we show the amplification of single large DNA. We used replication-cycle reaction (RCR), which makes it possible to amplify up to 1Mb large circular DNA with high fidelity. RCR was conducted in micro-sized w/o droplets. Even when single DNA was encapsulated in droplets, template DNA was amplified. We concluded that large single DNA was amplified by RCR.

2Pos104* Piezo1 はリンパ管弁形成過程におけるカルシウムシグナルに必要である
Piezo1 is required for calcium signaling during lymphatic valve morphogenesis

Hiroki Katsuta[1,2], Keiko Nonomura[2], Akemi Kanie[2], Takaki Miyata[1], Toshihiko Fujimori[2] ([1]*Grad. Sch. Med. Nagoya Univ. Cell Biol.*, [2]*NIBB Embryology*)

Lymphatic system is important for fluid circulation in the body and valves in the vessels are essential to prevent back flow. Previous studies have shown that Piezo1, one of mechanosensitive channels, is required for valve morphogenesis. However, its expression and activation patterns remain unclear, especially *in vivo*. By using Piezo1-tdTomato mice, we found that Piezo1 expression was high at valve-forming regions (VFRs) from early stages of valve morphogenesis. We also utilized GCaMP3 transgenic mice to monitor Ca signal. GCaMP3 fluorescence was high at VFRs, which was significantly impaired in endothelial specific Piezo1 cKO mice. From these results, it was suggested that site-specific calcium signaling by Piezo1 may be important for lymphatic valve morphogenesis.

2Pos105 血管新生における血管内皮細胞の往復運動
Linear reciprocating movement of vascular endothelial cells in angiogenesis

Naoko Takubo[1], Kazuaki Naemura[2], Ryo Yoshida[3], Terumasa Tokunaga[4], Hiroki Kurihara[2] ([1]*Isotope Science Center, Univ. Tokyo*, [2]*Grad. Sch. Med., Univ. Tokyo*, [3]*Inst. Statistical Mathematics*, [4]*Faculty of Computer Science and Systems Engineering, Kyushu Inst. Tec.*)

Vascular endothelial cells (ECs) in angiogenesis exhibit inhomogeneous collective migration called “cell mixing”. However, how ECs collectively form complex vessels remains largely unknown. To uncover hidden laws of integration driving angiogenic morphogenesis, we analyzed EC behaviors in an in vitro angiogenic sprouting assay using mouse aortic explants. Time-lapse imaging of sprouts showed U-turn movement of individual cells in inhomogeneous collective migration of ECs. The U-turn movement can be summarized that an EC persistently moves toward one end of a branch, and likewise return to the other end triggered by a following cell’s proximity. That is a linear reciprocating movement of individual ECs in angiogenesis.

2Pos106 ドリフトのあるランダム運動がヒトの胚葉形成には必要である
Brownian motion with drift is essential for forming human germ layers

Kenshiro Maruyama, Ryo Kobayashi, Haru Hikita, Tadashi Tsubone, **Kiyoshi Ohnuma** (*Nagaoka University of Technology*)

To understand the dynamics of forming the human body, we utilize human pluripotent stem cells (iPSCs), which roughly correspond to the early embryo. We focused on the dynamics of mesoderm and endoderm (future muscle, gut, etc.) layer formation during the gastrulation stage. Single-cell dynamics of the iPSCs derived mesoderm and endoderm cells were measured by time-lapse imaging. A random walk analysis of these cells showed that the migration modes are both random. Computer simulation based on the measured dynamics showed that not only random movement but also drift (external physical force or chemotaxis) was required for forming homogeneous discoidal germ layers of the human. These results are consistent with the other amniotes development.

2Pos107 確率的な細胞たちが協調して正確な大きさの体節を作る仕組み ～分節時計によるノイズキャンセル機構～
Noise-resistant developmental reproducibility in vertebrate somite formation

Naoki Honda[1,2], Ryutaro Akiyama[2], Dini WK Sari[2], Bessho Yasumasa[2], Takaaki Matsui[1,2] ([1]*Grad. Sch. Biostudies., Kyoto Univ.*, [2]*NAIST*)

The segmentation clock has been widely considered vital for somite formation, because clock-deficient embryos display severe segmental defects. However, irregular somites are still formed, suggesting that the clock is not required for somite formation itself but rather endows it with developmental reproducibility. Thus, the following questions arose: How do irregular somites emerge in a clock-independent manner? How is the irregularity reduced in the presence of the clock? To address these questions, we developed a computational model of somitogenesis. We then clarified that the intrinsic noise induces irregular somite formation in the absence of the clock, and that the clock plays an important role in suppressing the noise effect for reproducible somite formation.

2Pos108 エネルギー地形アプローチに基づく組織形態形成の多様性と安定性の探求
Exploring variety and robustness in tissue morphogenesis based on energy landscape approach

Yoshitaka Kameo[1,2,3], Hironori Takeda[2], Taiji Adachi[1,2,3] ([1]*IFLMS, Kyoto Univ*, [2]*Grad Sch Eng, Kyoto Univ*, [3]*Grad Sch Bio, Kyoto Univ*)

Tissue morphogenesis proceeds under the effects of mechanical forces generated by multi-cellular activities such as proliferation and constriction. Although the underlying molecular and cellular behaviors have been identified, the regulatory mechanism of how various tissue morphologies are robustly formed even under thermal fluctuation is far from understood. In this study, we proposed a novel approach to capture the overall view of mechanically admissible morphogenesis by describing the landscape of strain energy accumulated in the growing tissue. This approach enables us to understand the variety and robustness in tissue morphogenesis from a mechanical viewpoint by investigating the behavior of local stable states on the energy landscape.

2Pos109 三次元粒子画像流速測定法によるノード流の解析
Analysis of nodal flow by three-dimensional particle image velocimetry

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In mammalian development, a leftward flow (nodal flow) generated by nodal cilia determines future asymmetry. The mechanism how the embryo detects the flow is still in question. The mechanosensing model, a major hypothesis, proposes nodal cilia function as mechanosensor in addition to as flow generator. Since the left and the right side of the node surface are exposed to the flow, the sensor cilia need to have anisotropy, or the flow velocity to be different between the two sides. To test the second possibility, we observed three-dimensional flow pattern of nodal flow using a high-speed light-sheet microscope with an electrically tunable lens. Images of fluorescent beads in the node cavity were acquired at ~10 volume/sec, then analyzed by a particle image velocimetry.

2Pos110 Mechanical stress-responsive membrane remodeling in muscle cells

Kenshiro Fujise, Hiroshi Yamada, Kohji Takei, **Tetsuya Takeda** (*Okayama Univ. Grad. Sch. Med. Dent. Pharm. Sci.*)

T-tubules are membrane invagination of muscle cells essential for E-C coupling. Dysfunction of T-tubules induces muscle weakness and atrophy leading to muscle disorders such as myopathy. Centronuclear myopathy (CNM) is a congenital myopathy caused by mutations in membrane remodeling proteins Dynamin-2 and BIN1. Skeletal muscles are subject to constant mechanical stresses due to repeated contraction and relaxation. However, CNM pathogenesis caused by defective membrane remodeling under mechanical stresses remained to be elucidated. In this study, we present our recent findings about mechanical stress-response of the T-tubule-like membrane invagination induced by BIN1 overexpression in mouse myoblast C2C12 cells and discuss about its contribution to the CNM pathogenesis.

2Pos111 無機ポリリン酸存在下でのアクチン重合
Polymerization of actin molecules in the presence of inorganic polyphosphate

Koji Ito, Kuniyuki Hatori (*Grad. Sch. Sci. Eng. Yamagata Univ.*)

Inorganic polyphosphates are known to increase a water-holding capacity of muscle homogenates. We have reported that actomyosin can hydrolyze triphosphate and the reaction is uncoupled to the motility (Ito et al., BBA 1866, 1224-1231, 2018). To examine the effect of polyphosphate on actin polymerization, here we measured the fluorescence intensity of pyrene bound to actin monomers during their filament formation. The apparent binding rate constant tended to be higher in the presence of mono-phosphate or pyrophosphate than in the presence of ATP, whereas tri- or tetra-phosphate decreased the rate constant.

2Pos112 Visualization of Ca2+ regulated structural change in muscle thin filament by cryoEM

Yurika Yamada, Keiichi Namba, Takashi Fujii (*Grad. Sch. of Frontier Biosci., Osaka Univ.*)

Muscle contraction is driven by cyclic interactions of myosin in the thick filament with actin in the thin filament composed of actin, tropomyosin (Tm) and troponin complex (TnC, TnI, TnT). It is thought that the binding of Ca2+ released from sarcoplasmic reticulum to TnC triggers a conformational change of Tm on the actin filament to allow the actin-myosin interaction. To understand this regulatory mechanism, high resolution structures of the thin filament in different states are necessary. We solved the structures of entire thin filament with Tm and Tn by cryoEM in the absence and presence of Ca2+ and revealed two distinct arrangements of Tn and Tm on the actin filament. We will discuss the regulatory mechanism of muscle contraction by comparing the two structures.

2Pos113 Fhod3 と cMyBP-C による心筋サルコメアの形成および維持機構
Mechanism of construction and maintenance of cardiac sarcomeres by Fhod3 and cMyBP-C

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Fhod3, a formin homology protein, and cMyBP-C, a cardiomyopathy-related protein, interact with each other in C-zone of the heart sarcomere and is known to regulate the formation and maintenance of cardiac sarcomeres, but the detail is unknown. Here, by plastic-embedded thin sections of cardiac sarcomeres and electron tomography, we elucidate the 3D-structure of cardiac sarcomeres of wild type and the mutant where cMyBP-C was deleted and/or Fhod3 was over-expressed. Then, we found that the actin had variety in length. Furthermore, cardiac sarcomere lattices and Z-lines were also disordered. These could be probably induced by abnormal regulation of actin polymerization due to the loss of the interaction. We try to visualize their direct interactions in wild type etc.

2Pos114* DNA オリガミを用いたナノバネ結合心筋ミオシンフィラメントの 1 分子解析
Single molecule analysis of DNA origami-based cardiac myosin filaments attached with Nanospring

Hiroki Fukunaga[1], Masashi Ohmachi[2], Keisuke Fujita[2], Keigo Ikezaki[3], Toshio Yanagida[1,2], Mitsuhiro Iwaki[1,2] ([1]*FBS, Univ. Osaka*, [2]*BDR, Riken*, [3]*Grad. Sch. Sci., Univ. Tokyo*)

Muscle contraction is generated by a coordinated motion of myosin motors in thick filaments. While mechanical motion of isolated single myosins is well characterized, how individual myosins in an assembly are coordinated together is unclear. Previously we have developed artificial myosin filaments composed of DNA origami and recombinant human muscle myosins, which gives us an analytical platform to clarify both single and ensemble myosin II dynamics. Here, we attached human cardiac α and β myosins at controlled position on the platform and also linked with Nanospring to apply force to the filaments, enabling estimation of mechanical properties like force-velocity and power-force relation.In this meeting, we will present the current progress of our experiments.

2Pos115* 細胞質ダイニンの運動方向性を左右するアミノ酸の同定
Key residues on cytoplasmic dynein for asymmetric unbinding from microtubule

Shintaroh Kubo[1], Tomohiro Shima[2], Takahide Kon[3], Shoji Takada[1] ([1]*Grad. Sch. Sci., Univ. Kyoto*, [2]*Grad. Sch. Sci., Tokyo Univ.*, [3]*Grad. Sch. Sci., Univ. Osaka*)

Cytoplasmic dynein is a two-headed molecular motor that moves to the minus end of microtubule (MT). The unbinding rate of the motor from MT shows directional dependency. This asymmetric unbinding has been proposed to be critical for unidirectional movement of dynein, but the molecular basis for the property remains obscure. Here, using MD simulation, we clarified the key residues for the property. In vitro experiments showed that point mutations of the residues indeed abolished dynein's directionality. Our results suggest the molecular basis of asymmetric unbinding and its importance on dynein movement.

2Pos116* 好熱菌由来 F_1-ATPase の至適生育温度における力学的仕事の測定
The Measurement of Mechanical Work of Thermophilic F_1-ATPase at the Optimum Growth Temperature

Tomoaki Okaniwa[1], Yohei Nakayama[2], Naoya Terahara[1], Eiro Muneyuki[1] ([1]*Dept. Phys., Graduate School of Science and Engineering, Chuo Univ.*, [2]*Dept. Appl. Phys., Graduate School of Engineering, Tohoku University*)

We measure mechanical work of thermophilic F_1-ATPase (TF_1) in the single molecule experiment at the optimum growth temperature, 65°C. It has been already found that the thermodynamic efficiency in converting free energy released by ATP hydrolysis to mechanical work by TF_1 reaches nearly 100% at 25°C [PNAS. 108, 17951(2011)]; however, the robustness of the high thermodynamic efficiency to the environmental condition has been elusive. Thus, we enable long time rotation assay of TF_1 at high temperature such as 65°C and estimate the maximum work which is generated by TF_1 against external torque in order to elucidate whether TF_1 retains high thermodynamic efficiency at the optimum growth temperature.

2Pos117* 中間鎖をアンカーとした新しい運動アッセイ法によるクラミドモナス軸糸ダイニン集団の運動特性の計測
Collective motility of Chlamydomonas outer arm dynein measured using its intermediate chain as a scaffold for motility assays

Yuka Matsuda[1], Akane Furuta[2], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2], Ken'ya Furuta[2] ([1]*Grad. Sch. Sci., Univ. Hyogo*, [2]*Adv. ICT Res. Inst., NICT*)

Flagellar beating is generated by thousands of dynein motors periodically distributed along the axonemal microtubule (MT). To understand how the individual dynein motors contribute to the organized beating waveforms, we set out to build a simplified axoneme-like system using DNA nanotubes as a template. As a first step, we engineered the one of dynein subcomponents, an intermediate chain 2 of outer arm dynein (OAD) for fusing with a His-tag and a SNAP-tag, which allows affinity purification of OAD using a Ni column and conjugation to DNA nanotubes via SNAP-tag. Purified OAD showed robust MT gliding activity and load-dependent MT-binding behavior when applied force by an optical trap. We demonstrated the collective force generation by DNA-templated assembly of OADs.

2Pos118 The combination of high-speed atomic force microscopy and X-ray crystallography reveals rotary catalysis of a shaftless V1 motor

Shintaro Maruyama[1], Motonori Imamura[2], Takayuki Uchihashi[3,4], Kazuya Nakamoto[1], Kenji Mizutani[5], Lica Fabiana Imai[1], Kano Suzuki[1], Yoshiko Ishizuka-Katsura[6], Tomomi Someya-Kimura[6], Mikako Shirouzu[1,6,7], Ichiro Yamato[1,7], Toshio Ando[2], Takeshi Murata[1,8] ([1]*Grad. Sch. Sci., Univ. Chiba*, [2]*WPI Nano Life Sci. Inst., Univ. Kanazawa*, [3]*JST, CREST*, [4]*Dep. Phys., Univ. Nagoya*, [5]*Grad. Sch. Med. Life. Sci., Univ. Yokohama*, [6]*DSSB, RIKEN*, [7]*Ind. Sci. Tokyo Univ. Sci.*, [8]*PREST, JST*)

V1-ATPase is an ATP-driven rotary motor that is composed of a ring-shaped A3B3 complex and a central DF shaft. An A3B3 stator ring sequentially undergoes conformational changes upon ATP hydrolysis to rotate the central shaft DF unidirectionally. To reveal the rotation mechanism of V1-ATPase, first, we used high speed atomic force microscopy to confirm that the asymmetrical structure of the cysteine cross-linked A3B3 complex (cysA3B3) propagated different conformational states in the anticlockwise direction. Second, we determined three kinds of crystal structures of the cysA3B3 as the snapshots of each steps in the rotation. These findings further the understanding of the principle of the unidirectional rotation mechanism of rotary motors.

2Pos119* Measurement of force generation by dynein ensemble on a doublet microtubule obtained from sperm flagella

Takashi Fujiwara[1], Chikako Shingyoji[1], Hideo Higuchi[2] ([1]*Dept. Biol. Sci., Grad. Sch. Sci., Univ. Tokyo*, [2]*Dept. Phys., Grad. Sch. Sci., Univ. Tokyo*)

Cyclical beating of eukaryotic flagella is caused by orchestrated activity of dynein molecules to produce sliding among nine doublet microtubules (DMT). To understand behavior of dyneins in flagella, we measured force generation of dynein ensemble on a DMT by laser tweezers. Individual DMTs on which dynein arrays had been arranged were obtained by disintegration of protease-treated axonemes. Polymerized microtubule that bound to a trapped bead was manipulated to interact with hundreds of dyneins on a DMT. Dyneins generated force of 5-6 pN in average, which is equivalent to the force of a single dynein molecule. This result indicates that only ~1% of dynein on a DMT is possibly active and the remaining dyneins may be inactive or weakly bind to the microtubule.

2Pos120* 1 分子計測による腸内連鎖球菌由来の回転分子モーター V_1-ATPase の化学力学共役機構の解明
Chemo-mechanical coupling scheme of rotary molecular motor *Enterococcus hirae* V_1-ATPase revealed by single-molecule analysis

Tatsuya Iida[1,2], Yoshihiro Minagawa[3], Hiroshi Ueno[3], Fumihiro Kawai[4], Takeshi Murata[5], Ryota Iino[1,2] ([1]*SOKENDAI (The Grad. Univ. for Adv. Stud.)*, [2]*Inst. for Mol. Sci.*, [3]*The Univ. of Tokyo*, [4]*Yamagata Univ.*, [5]*Chiba Univ.*)

A rotary molecular motor V_1-ATPase (V_1) converts chemical energy of ATP into mechanical rotation. To understand chemo-mechanical coupling scheme, we conducted single-molecule analysis of *E. hirae* V_1 (EhV_1). We found that 120° steps per ATP hydrolysis were divided into 40° and 80° substeps which are triggered by ATP binding and ADP release, respectively. We propose a model of chemo-mechanical coupling scheme of EhV_1, combined with previous structural information. In our model, ATP cleavage and product Pi release occur at 240° when ATP binds at 0°, then product ADP is released at 280°. The scheme is largely different from those of F_1-ATPases. This difference may be relevant to physiological functions of V-ATPase and F-ATP synthase, active ion transport and ATP synthesis.

2Pos121 (2SDA-7) Dynamic energy landscape of a linear motor chitinase from single-particle tracking trajectories

Kei-ichi Okazaki, Akihiko Nakamura, Ryota Iino (*Institute for Molecular Science*)

Single-molecule experiments have been used to investigate mechanism of biomolecular motors by visualizing their motions. In this study, we estimate an underlying diffusion model from observed single-molecule trajectories. To deal with nonequilibrium trajectories driven by chemical energy consumed by biomolecular motors, we develop a novel framework based on hidden Markov model, which considers switching among multiple energy surfaces depending on chemical states of the motors. The method is applied to single-molecule trajectories of processive chitinase, a linear motor that moves with hydrolysis energy of single chitin chain. The dynamic free energy landscape underlying the burnt-bridge Brownian ratchet mechanism of chitinase is revealed.

2Pos122 アクチンの pH 依存的な荷電状態と分子間相互作用
pH-dependent charge-state and intermolecular interaction of actin

Jun Ohnuki, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

Actin filaments can change the interaction with actin-binding proteins (e.g., cofilin, myosin) in response to the changes in the bound nucleotide, tension, and pH. We previously found the dielectric allostery and piezoelectric allostery in actin where the nucleotide-state change and the applied tension, respectively, induce the charge displacement in actin and regulate the intermolecular interactions. The decrease in pH stabilizes the filament and accelerates the cofilin binding although the underlying physical mechanism is unclear. By conducting constant pH molecular dynamics simulation, we investigate the pH-dependent charge-state of actin and its effect on the actin-actin and the actin-cofilin binding.

2Pos123 枯草菌べん毛モーターの回転ステップ解析
Step analysis of the *Bacillus* Na^+-driven flagellar motor

Naoya Terahara[1,2], Miku Yoh[3], Eiro Muneyuki[1], Keiichi Namba[2,4], Tohru Minamino[2] ([1]*Dept. Phys., Chuo Univ.*, [2]*Grad. Sch. Frontier Biosci., Osaka Univ.*, [3]*Fac. Human Life Sci., Doshisha Women's Col.*, [4]*BDR/RSC, Riken*)

The bacterial flagellar motor consists of a rotor and a dozen stators and is driven by ion motive force across the cytoplasmic membrane. The stator acts as a transmembrane ion channel to couple the ion flow through the channel with torque generation. Although the motor seems to be continuously rotating, 26 steps per revolution have been observed. However, since about ten stators are simultaneously incorporated into the motor, it is difficult to define the elementary process through stator-rotor interactions coupled with the ion flow. To clarify the energy coupling mechanism of the *Bacillus* Na^+-driven MotPS motor, we performed high-resolution rotation measurements of the motor with one or two active stators and will discuss Na^+-coupled stepping mechanism of the motor.

2Pos124 アクチン繊維の集団運動により形成されるベルトパターンはリング状に変化する
Transformation of belt-like to ring patterns of a quasi-concentrated solution of F-actin driven by myosin-coated surface

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A quasi-concentrated solution of filamentous actin was known to form belt-like patterns of about 20 μm width when driven by HMM-coated glass with a strict plane (ref. 1). Their width and the curvature were found to be varied according to their solution conditions such as ionic strength or temperature. In this report, we will report that the patterns were transformed into the assembly of rings with about 60 μm diameter. Considering that the protein molecules contributing to the pattern formation are only actin and myosin, the nature for ring-pattern formation may be the intrinsic property of these proteins. The fact may insinuate the relation to some biological process. (ref. 1. Kentaro Ozawa, et al. Biophysics and physicobiology 16 (2019): 1-8.)

2Pos125 Myosin II decreases the connectivity of an actin network using two different mechanisms depended on concentration of crosslinking protein

Kyohei Matsuda[1], Takuya Kobayashi[1,2], Mitsuhiro Sugawa[1], Masahiko Yamagishi[1], Yoko Y. Toyoshima[1], Junichiro Yajima[1] ([1]*Department of Life Sciences, Graduate School of Arts and Sciences, The University of Tokyo*, [2]*Juntendo University*)

Myosin II enables an actin network with actin crosslinking protein (ACP) to perform mechanical work such as contraction. However, it remains unclear how the mechanical properties of myosin contract ACP-actin network (AAN) into clusters. Here, we added myosin and ATP to AAN, and observed the morphology and the contractile motion of the network. We found that the contraction velocity was fast at high myosin or low ATP. The cluster size was large at high myosin and was small at low ATP. Further, we observed the actin fragmentation in AAN at low ATP. Our findings indicate that myosin can decrease the connectivity of AAN using two different mechanisms that one is the dissociation of ACP at low ACP, and the other is actin fragmentation at high ACP.

2Pos126 Developing the detection system of the conformational change in rotating flagellar motor by single motor FRET

Takuma Nakagawa, Tatsuya Yamakoshi, Yong-Suk Che, Akihiko Ishijima, **Hajikme Fukuoka** (*Grad. Sch. Frontier Biosci., Osaka Univ.*)

Flagellar motor of *E. coli* is a rotary motor and its rotational direction switches between CCW to CW directions. In this study, to understand the process of conformational change during rotational switching via FRET in rotating motor, we developed CFP-fusion of FliM and YFP-fusion of FliN. We succeeded in the detection of FRET in a single rotating motor by analyzing the ratio of intensity between CFP and YFP channels for the fluorescent spot at the rotational center of tethered cell. Moreover, FRET appeared to occur efficiently between C-terminus of FliM and N-terminus of FliN, indicating these regions would be close to each other. We will discuss about the conformation of rotating motor based on results via simultaneous measurement of FRET and rotational switching.

2Pos127 ダイニン・微小管・DNA 折り紙複合体の振動的運動
Oscillatory movement of the dynein-microtubule complex crosslinked with DNA-origami

Shimaa A. Abdellatef[1], Hisashi Tadakuma[2], Yuichi Kondo[3], Kangmin Yan[1], Hideo Higuchi[3], **Keiko Hirose**[1] ([1]*Biomed. Res. Inst., AIST*, [2]*IPR, Osaka Univ.*, [3]*Grad. Sch. Sci., Univ. Tokyo*)

Oscillation of cilia and flagella is thought to be caused by alternate activation of dynein molecules on the opposite sides of the axoneme, but it is not clear how switching of the activity occurs. Here we made a system that contains outer-arm dynein bound between two microtubules (MTs) in two different orientations, and crosslinked the MTs with DNA origami. In optical trapping experiments, the complex showed oscillatory displacement of the MT after photolysis of caged ATP. Both the forward and backward movement showed steps of ~7 nm, indicating that both are caused by active stepping of dynein. The results suggest that a system composed of only dynein, MTs, and flexible linkers has an intrinsic ability to self-regulate the activity and move in an oscillatory manner.

2Pos128 Ca2+濃度変化による真核生物鞭毛軸糸の構造変化の X 線繊維回折解析
Structural changes of Chlamydomonas and Ciona flagellar axonemes coupled with the change in [Ca2+] studied with X-ray fiber diffraction

Kazuhiro Oiwa[1], Hiroyuki Iwamoto[2], Kogiku Shiba[3], Kazuo Inaba[3], Hitoshi Sakakibara[1] ([1]*Adv. ICT Res. Inst., NICT*, [2]*JASRI*, [3]*Shimoda Marine Res. Cent. Univ. Tsukuba*)

The mechanism of waveform changes coupled with intracellular [Ca2+] found in Chlamydomonas and Ciona flagella has remained a long-standing unresolved issue. We have investigated this mechanism using the continuous shear-flow alignment of flagellar axonemes and X-ray fiber diffraction in the synchrotron radiation facility SPring-8, BL40XU. The diffraction pattern of Ciona sperm flagella was similar to that of Chlamydomonas but had some missing reflections. We explored the spatial arrangement of axonemal components under different [Ca2+]. In the low [Ca2+], the radial-spoke-originating 48-nm and 24-nm meridional reflections split each into the layer lines, suggesting changes in the helical symmetry of nine doublet microtubules in the axoneme.

2Pos129 新規繊毛ダイニン軽鎖 MOT7 の構造機能解析
Structural/functional analyses on MOT7, a novel light chain of ciliary dynein f/I1

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In this study, we have analyzed the “MOT7” protein in Chlamydomonas, an orthologue of the novel light chain of the ciliary dynein (DYBLUP) first identified in Ciona sperm. Chlamydomonas null mutant of MOT7 showed ciliary-waveform and swimming-velocity defects, and we confirmed that MOT7 is a light chain of the ciliary dynein “f/I1” that regulates ciliary movement in Chlamydomonas as well as in Ciona. From biochemical and structural analyses, we revealed that MOT7 possibly interacts both with motor domain(s) of dynein “f/I1” and “FAP44”, a component of the “f/I1-tether complex” which is another important regulator in cilia. We hypothesize that MOT7 may coordinate the activities of these regulators to generate the proper ciliary waveform.

2Pos130 全反射赤外分光法を用いた共役イオン結合による Na^+駆動型モーター固定子の構造変化の解明
The cation-induced structural changes in the Na^+-driven flagellar stator studied by ATR-FTIR

Masayo Iwaki[1], Tatsuro Nishikino[2], Hiroyuki Terashima[2], Michio Homma[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech.*, [2]*Nagoya Univ.*)

The flagellar motor of marine bacteria *Vibrio alginolyticus* is driven by a Na^+-gradient across the cytoplasmic membrane. The rotor-stator interactions coupled with ion translocation through the stator are essential for torque generation. In this study, to elucidate the mechanism of cation-translocation in the stator, the cation-induced molecular structural changes in the purified stator complex, PomA/PomB, were studied by ATR-FTIR spectroscopy. The cation-sensitive IR changes were measured in the WT PomA/PomB, as well as in the mutants, which impaired the motility and/or ion translocation. Based on the vibrational analysis of the IR data, cation-induced structural changes in PomA/PomB, which could lead the torque generation, will be discussed.

2Pos131 2 つのアゾベンゼンを持つフォトクロミック Eg5 阻害剤存在下における Eg5 活性の光制御機構の研究
Study on inhibitory mechanism of kinesin Eg5 with photochromic Eg5 inhibitor composed of two azobenzene

Kei Sadakane[1], Islam MD Alrazi[2], Kenichi Taii[2], Tomisin H. Ogunwa[3], Shinsaku Maruta[1,2] ([1]*Sci. & Eng., Soka Univ.*, [2]*Grad. Sch. Eng., Soka Univ.*, [3]*Grad. Sch. Fisheries and Environmental Sci., Nagasaki Univ.*)

Mitotic kinesin Eg5 has been expected as a target protein for the cancer therapy. It is known that there are several small molecules which exhibit specific potent inhibition for kinesin Eg5. Previously, we synthesized a novel photochromic Eg5 inhibitor composed of two azobenzene derivatives (BDPSB). BDPSB inhibited the function of Eg5 photo reversibly. Trans isomer of BDPSB exhibited potent inhibition on the ATPase and motor activities of Eg5. On the other hand, cis isomer showed almost no inhibition. In this study, we have performed kinetic study on the ATPase inhibition of Eg5 with BDPSB, mixed motor assay with conventional kinesin and in silico study. The preliminary results indicated that BDPSB performs as non-competitive inhibitor for kinesin Eg5.

2Pos132 結晶性キチン加水分解酵素は背水の陣で進むブラウニアンモーターである
Crystalline chitin hydrolase is a Burnt-bridge Brownian motor

Akihiko Nakamura[1,2], Kei-ichi Okazaki[1], Tadaomi Furuta[3], Minoru Sakurai[3], Ryota Iino[1,2] ([1]*Institute for Molecular Science*, [2]*SOKENDAI*, [3]*Tokyo Institute of Technology*)

SmChiA is a recently found as a liner motor protein which hydrolyses chitin molecular chain instead of ATP. Moving ability is important for degradation of stable chitin crystal, but moving mechanism is still not clear. Our single molecule observation results showed stepping size of SmChiA is 1.1 nm, and movement is highly biased forward. The ratio of kinetic rates of catalysis and backward step was similar to the forward stepping ratio. Additionally, X-ray crystal structure of SmChiA in sliding intermediate state was not different from the enzyme without chitin. These results mean SmChiA is a Burnt-bridge Brownian ratchet motor which moves forward by shortening the chain end to inhibit the backward movement.

2Pos133 プログラマブルな DNA オリガミによる微小管格子構造の制御
Defining microtubule lattice structure using programmable DNA-origami seeds

Daisuke Inoue, Franky Djutanta, Rizal Hariadi (*Biodesingn Institute, Arizona State Univ.*)

Microtubules (MTs) are hollow cylindrical biopolymers commonly composed of 13 parallel tubulin protofilaments (PFs) and play essential roles in many cellular processes. In living cells, the PF numbers and arrangements are tightly regulated by template protein complex. In contrast, the structure of in vitro MTs has been found to have a broad distribution of PF numbers and helicity. Consequently, single molecule studies over many decades have ignored the role of MT structures to their cellular functions. To bridge this gap between in vitro and in vivo experiments, well-defined assays are necessary, wherein the MT structures can be engineered. Here, we have developed a DNA origami seed to engineer precise number and spatial configuration of tubulins.

2Pos134 Plus-end directionality is present in the conserved catalytic motor core of kinesin-14 minus-end directed motors

Masahiko Yamagishi, Junichiro Yajima (*Dept. Life Sci., Grad. Arts & Sci., Univ. Tokyo*)

Molecular motor kinesins move along microtubules unidirectionally, which is essential for various cellular functions such as vesicle transport and cell division. However, the origin of directionality is not clear. Here we engineered monomer constructs of kinesin-14, the minus end directed kinesin, such that they could be anchored via either their N- or C-termini, leaving the opposite terminal regions mechanically disconnected from the surface. We find that kinesin-14 has intrinsic plus-end directionality derived of the conserved catalytic motor core like other plus end directed kinesins and that the proper function of the neck-helix region, outside of motor core, is required to achieve minus-end directionality of kinesin-14.

2Pos135 DNA オリガミの分子配置技術を用いたキネシン分子の協調性評価
Evaluating coordination between kinesin motors using DNA origami-based transport complex

Kodai Fukumoto[1], Yuya Miyazono[2], Hisashi Tadakuma[1], Yoshie Harada[1] ([1]*IPR, Osaka Univ.*, [2]*Grad. Sch. Front. Sci., Univ. Tokyo*)

Intracellular transport is central to the cellular phenomena, where multiple motors orchestrate their function. However, it remains elusive how individual kinesin contribute to the multiple-motor coordination and how the molecular layout (e.g. distance between individual motors) affect the cargo transport. To elucidate the coordination mechanism, it is useful to construct artificial transport complex. DNA origami provides an ideal scaffold to assemble motors in a spatially well-defined manner. Here, we have made DNA origami-based transport complex, where we integrated teams of kinesin containing SNAPf-tag and positively charged lysine-tag, and measured the movement on axonemes. This system may provide a framework to study the coordination behavior of motor proteins.

2Pos136 KIF1A/UNC-104 によるシナプス小胞前駆体輸送の数理モデル
Mathematic modeling of synaptic vesicle precursor transport by KIF1A/UNC-104

Ryo Sasaki[1], Ryota Shinagawa[1], Kazuo Sasaki[1], Shinsuke Niwa[2], Kumiko Hayashi[1,3] ([1]*Dep. Appl. Phys., Grad. Sch. of Eng., Tohoku Univ.*, [2]*FRIS, Tohoku Univ.*, [3]*JST, PRESTO, Tokyo, Japan*)

KIF1A/UNC-104 transports synaptic vesicle precursors (SVPs) along microtubule tracks in neurons. KIF1A/UNC-104 takes the autoinhibited form, when it does not haul the cargo vesicles not to consume ATP wastefully. This regulatory system controls the efficiency of the neuronal transport. Lack of the regulatory system leads to defects of synapse construction and neurological disease. In order to understand the defects of synapse construction and neurological disease from the autoinhibition mechanism, we performed numeral simulations of SVP transport model. We found that the change of the number of KIF1A /UNC-104 hauling an SVP, caused by the lack of the regulatory system, affected the efficiency of SVP transport.

2Pos137 遺伝性痙性対麻痺を引き起こす変異型ヒト KIF1A の運動特性
Motility characteristics of human KIF1A mutants in hippocampal neurons in relation to hereditary spastic paraplegia

Shiori Matsumoto[1], Kyoko Chiba[2], Shinsuke Niwa[3], Kumiko Hayashi[1,4] ([1]*Dep. Appl. Phys., Grad. Sch. of Eng., Tohoku Univ.*, [2]*Col. Biol. Sci., UC Davis*, [3]*FRIS, Tohoku Univ.*, [4]*PRESTO, JST*)

The kinesin 3 family member KIF1A is a molecular motor that transports synaptic vesicle precursors (SVPs) in the axons. In this study, we investigated the SVPs in mouse hippocampal neurons, carried by human KIF1A mutants, which cause hereditary spastic paraplegia. Previously, we found that the run-time, run-length, and velocity for the mutants were largely different from those for the wild-type (56th BSJ annual meeting, 2POS220). In order to understand the difference obtained in the hippocampal neurons, we perform in vitro single-molecule experiments, such as motility assay and optical trap experiments, which provide the detailed motility characteristics of KIF1A mutants.

2Pos138 キネシン 1 の連続的移動距離を決める要因の高速一分子観察
Determinant of the processivity of kinesin-1 as studied using high-speed single-molecule observations

Tsukasa Enomoto[1], Kohei Matsuzaki[2], Michio Tomishige[2] ([1]*Grad. Sch. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*)

Kinesin-1 is a motor protein that transports cargo inside the cells. Kinesin-1 has been shown to take ~100 steps on average before dissociating from microtubule, but it is still unknown how the run length is determined. Here, to elucidate the underlying mechanism for processivity, we used a heterodimer composed of a wild-type head and a mutant head that has a defect in ADP dissociation, and observed the motion using a high-speed dark-field microscopy. We found that the conformation of the heterodimer just prior to dissociation was mostly that the mutant head is in the leading and the wild-type head is in the trailing. This result suggests that kinesin dissociates from microtubule when the trailing head hydrolyses ATP before the leading head releases ADP.

2Pos139* 集団運動する神経幹細胞で測定された牽引力の、細胞配向場によるモデリング
Traction Force of Neural Stem Cells under Collective Migration was Modeled using the Orientation Field of Cell Alignment

Masahito Uwamichi[1], Kyogo Kawaguchi[2], Masaki Sano[3,4] ([1]*Dept. of Phys, Univ. of Tokyo*, [2]*RIKEN BDR*, [3]*IIAS*, [4]*TOKYO COLLEGE*)

Collective migration of spindle-shaped cells is observed as phenomena such as rostral migratory stream and cancer invasion after epithelial-mesenchymal transition. A phenomenological model from active matter field has been proposed to simply describe these collective migrations using its orientation field but is still to be checked by experiments. We observed the orientation field and measured the traction force in adhesive culture system of neural stem cells, to measure the active stress parameter which connects the orientation and the traction force in the model. The suitability of the model was also checked, and the result suggested the possibility to understand the mechanics of spindle-shaped cells from the viewpoint of cell orientation.

2Pos140* 細胞集団の 3D 自己組織化: 高分子溶液の水／水ミクロ相分離の活用
Self-Generating 3D Cellular Assembly: Aqueous/Aqueous Micro Droplet as a Non-invasive Scaffold

Ritsuki Ito, Toshifumi Kishimoto, Takahiro Kenmotsu, Koichiro Sadakane, Kenichi Yoshikawa (*Graduate School of Life Medical Science, Doshisha University*)

In cell biology as well as in regenerative medicine, fabrication of 3D cellular assembly is getting important. Artificial scaffolds such as gel and solid substrate have been usually applied. However, these scaffolds are possibly xenobiotics/invasive. Here, we will report the utilization of aqueous/aqueous micro-phase separation to generate 3D cellular assembly in a desired configuration. Owe to the minimum contribution of mixing entropy in aqueous solution with multiple polymers, phase-separation is caused. It is shown that coexisting charged biomacromolecule such as collagen exhibits the effect to stabilize the microdroplets. Under such experimental conditions, we have succeeded in constructing 3D cellular assembly with the microdroplet.

2Pos141* ナノ粒子を用いた細胞内局所加熱に対する細胞の応答
Responses of cells to local heating in cells using a nanoparticle

Hideaki Ota, Hideo Higuchi (*Phys., Grad. Sci., Univ. Tokyo*)

Cell function and motility depend much on temperature. Here, to understand relation between cells and localized high temperature in cells, we imaged the response of the cell under phase contrast microscope at the local temperature jump. Temperature was jumped up locally to about 57 °C by irradiating magnet nanoparticle (300 nm in diameter) in the cell with focusing infrared laser (wavelength of 1064 nm). By the temperature jump, (1) the most of vesicle transport in the cell stopped suddenly, (2) black circles at pseudopodia emerged and (3) the cell shrunk toward a heated nanoparticle. Result (1) and (2) often appeared together, so there is a deep relation between the two responses.

2Pos142* Quantitation of cell shape by machine learning

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Cell shape is closely related with many biological functions and changes depending on the environmental conditions. In spite of its importance, however, how cell shape is regulated is still unclear. The quantitation of cell shape is critical for revealing the regulation of cell shape and the relationship between shape and biological functions. Although many attempts have been made on quantitation of cell shapes, it is poorly understood how to quantitate the cell shape. In this study, we propose a novel method to quantitate cell shape with the use of Variational Auto Encoder (VAE). By using VAE, we extract feature values of any kinds of cell shape and construct a low dimensional space by feature values. We draw and observe the trajectory of cell shape in that space.

2Pos143* 細胞接着形態振動を駆動するグラフトポリマー層の垂直/水平変形性とその相関解析
Dual characterizations of vertical/lateral deformation of grafted-polymer layer driving cell-shape oscillation

Sayaka Masaike[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

The shape of mesenchymal stem cells (MSCs) influences on the differentiation: e.g., spread shape; osteogenic, contracted shape; adipogenic lineage. To maintain the stemness of MSCs, we have developed a grafted-polymer substrate that repeatedly switches the cell shape between them. However, the mechanism of shape switching is unclear yet. In this study, we investigated effect of both vertical and lateral deformation of the grafted-polymer on the cell adhesion behaviors. We report that each of the vertical and lateral deformation of grafted-polymer layer exhibit a certain threshold to modulate stability of focal adhesions and to alter cell shape, and discuss about maintenance of stemness of MSCs through nucleocytoplasmic shuttling of mechanotransducers such as YAP.

2Pos144* 鍵アミノ酸残基を含む領域に変異をもつべん毛繊維の in vitro 多型変換実験
In vitro polymorphic transformation of flagellar filaments with mutations in a domain including the key amino acids

Shiori Hirose[1], Hidetoshi Tomaru[1], Yuuka Sashida[1], Yuka Kobayashi[1], Kana Horiguchi[1], Mikako Tsubaki[1], Fumio Hayashi[2], Kenji Oosawa[1] ([1]*Gunma Univ, Grad. Sch. Sci. Technol*, [2]*Gunma Univ, Ctr. Instr. Anal*)

During tumbling of Salmonella, the helical shape of flagellar filament changes and this is called flagellar polymorphic transformation. We proposed amino acids involved in this polymorphic transformation as the key residues. Among mutants with a second mutation in them, flagellar filaments of A427T transformed from curly to coil to curly within a narrow pH range from 10.3 to 11.4. It is suggested that, lysine is responsible for from curly to coil polymorphic transformation at pH 10.4 and that for from coil to curly at pH11.3 maybe arginine related. Results from, Q117A and R431K also will be discussed.

2Pos145* マイコプラズマニューモニエの走流性
Rheotaxis in *Mycoplasma pneumoniae*

Yoshiki Kabata, Daisuke Nakane, Takayuki Nishizaka (*Department of Physics, Gakushuin University*)

Bacterial cell motility is controlled in response to a changing environment. A small parasitic bacterium, *Mycoplasma pneumoniae*, is capable to adhere and move over host cell surface, but there was no report so far about the taxis in their motility. Here we found that they have positive rheotaxis in a flow-controlled chamber; the cell re-oriented and moved upstream in a fluid flow. The gliding speed decreased linearly with a slope of 0.06 in a range of flow rate at 1.1-4.2 mm s^{-1}. This study provides the insight into how cell moves under a flow from mucociliary clearance in respiratory tract.

2Pos146* 細胞内で LLPS 現象を観察する簡単な方法の開発
Development of a simple method to observe LLPS in cells

Chaieun Kim[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo.*, [2]*PRESTO, JST*)

Liquid-liquid phase separation (LLPS) has various functions including stress granule formation in cells. Previous studies have revealed that aggregations of proteins with low complexity domains provoke LLPS in vitro. However, it was difficult to investigate LLPS in cells. In this study, we solved this problem by utilizing TIA1, which has self-assembly domains. First, it was confirmed that fluid and reversible granules were formed only by overexpressing TIA1-GFP. Furthermore, these cells showed significantly lower level of eIF2α phosphorylation than physiologically stressed cells. These results showed that aggregation of TIA1 underwent LLPS in cells. In the future, this method is expected to be applied to the study of the initiation mechanism of LLPS in cells.

2Pos147 (2SCP-5) Morphodynamic feature space of migrating cells

Daisuke Imoto[1], Nen Saito[2], **Satoshi Sawai**[1,3] ([1]*Graduate School of Arts and Sciences, University of Tokyo*, [2]*Universal Biology Institute, Graduate School of Science, University of Tokyo*, [3]*Research Center for Complex Systems Biology, University of Tokyo*)

We will introduce a minimalistic model that describes the essential regulatory logics underlying complex morphology of amoeboid cells. The proposed model recapitulates the overall cell forms observed in freely migrating Dictyostelium, neutrophils and fish keratocyte. Similarities between the simulated data and real cell data were assessed by feature extraction by deep learning. Our numerical analyses suggest importance of speed of membrane protrusion and the total amount of key positive regulators of membrane protrusion. Theoretical analysis will be presented along with demonstration of applicability of our approach to analyzing the migratory response in chemotattractant field and cell-cell adhesion.

2Pos148 バクテリア集団運動の揺らぎと応答の測定
Measurement of fluctuation and response of bacterial collective motion

Tatsuro Kai, Takahiro Abe, Shuichi Nakamura, Seishi Kudo, Shoichi Toyabe (*Appl. Phys., Tohoku Univ.*)

Bacteria realize taxis by repeating “run” and “tumbling” motions. We discuss how the interaction among bacteria modifies the swimming behavior, especially in terms of taxis. We first show that the diffusion of each Salmonella cell depends on the cell density and has a peak at a specific density. This suggests that the cellular interaction affects the direction-change frequency. We further examined the effect of collective motion on the taxis behavior under a local temperature gradient created by LASER irradiation. A remarkable observation is that the taxis speed increased with the cell density, suggesting the enhancement of the taxis by cellular interaction. We also observed that bacteria aggregated to a donut-like shape around the high-temperature region.

2Pos149 細胞運動のメカノケミカルモデル
Mechanochemical modelling of crawling cells

Mitsusuke Tarama[1], Kenji Mori[2], Ryoichi Yamamoto[2,3] ([1]*RIKEN BDR*, [2]*Dep Chem Eng, Kyoto Univ*, [3]*IIS, Univ Tokyo*)

Biological cells move by converting chemical reactions into mechanical forces. Since the force that a cell produces should vanish in total due to the law of action and reaction, it is not trivial how it can migrate spontaneously. A typical mechanism of cell crawling consists of four processes: 1. protrusion of the leading edge, 2. adhesion of the leading edge to the substrate underneath, 3. deadhesion of the trailing edge, and 4. contraction of the trailing edge. By introducing a simple mechanical model, we investigate how a net migration is achieved from these four processes under the force-free condition. Further, we extend the model to a mechanochemical model that combines intracellular chemical reactions and mechanics.

2Pos150 Quantitative Analysis of Signal-dependent Cell Cycle Regulation

Kyoichi Ebata, Hiroaki Imoto, Sawa Yamashiro, Mariko Okada (*IPR, Osaka Univ.*)

Cell is a complex regulatory system that recognizes and processes external cues to determine its phenotype. Although growth factor (GF) signaling and cell cycle have been well studied, the quantitative regulatory mechanism of signal-dependent cell cycle is still unclear. In this research, we simultaneously analyzed time-course of 8 cell cycle regulators and 4 signaling mediators in order to unveil the dynamic control of cell cycle. Our experimental and computational analyses suggested the signaling dynamics affecting timing and amplitudes of each cell cycle regulators. The analyses also indicated the presence of known and unknown cell cycle regulatory mechanisms.

2Pos151 飢餓状態への過渡期における大腸菌集団の束状凝集と一細胞形態
Bundle structure and single-cell morphology in E. coli populations during transient to a starvation condition

Takuro Shimaya[1], Reiko Okura[2], Yuichi Wakamoto[2], Kazumasa A. Takeuchi[1,3] ([1]*Dept. of Phys., Univ. Tokyo*, [2]*Dept. of Basic Sci., Univ. Tokyo*, [3]*Dept. of Phys., Tokyo Tech*)

To investigate how spatial structure of dense bacterial populations react against changes in the culture condition, we develop a new microfluidic device, which we call “extensive microperfusion system”. This device allows us to observe dense bacterial suspensions in two-dimensional space under a uniform environment. We then continuously observe morphological response of E. coli populations during starvation. We find that each cell aggregates and forms bundle structure after we remove nutrient from media. Simultaneously, the normalized cell-length distribution gradually sharpens. In the presentation, we will discuss the relationship between the bundle structure and the cell-length distribution and also show several observations obtained by the new system.

2Pos152 Adaptability and robustness of cell migration realized by size-dependent polarity dynamics

Akihiko Nakajima[1,2], Motohiko Ishida[2], Satoshi Sawai[2,3] ([1]*Dept. Gen. Sys. Studies, Grad. Sch. Arts & Sci., Univ. Tokyo*, [2]*Res. Cent. Comp. Sys. Biol., Univ. Tokyo*, [3]*Dept. Basic Sci, Grad. Sch. Arts & Sci., Univ. Tokyo*)

Although it has been reported that migratory patterns of immune cells and cancer invasiveness are highly heterogeneous in vivo, the mechanistic basis of the various migratory phenotypes remains almost unexplored. Here, we developed a microfluidic system with a parallel array of linear channels of several micrometers wide that allows high-resolution image acquisition from several hundred isolated cells in a single run. From the high-throughput imaging analysis, it was found that there were several migration modes in the population and the stability of cell polarity was dependent on cell size. We will discuss how the balance of the leading and trailing edge activities results in the adaptability and robustness of cell polarity.

2Pos153 3 次元毛細管構造のトポロジーが血管内皮細胞のシート展開の振る舞いを決定する
Topology of three-dimensional capillary structure determines blood vein sheet extension behavior

Kento Iida[1], Hiromichi Hashimoto[1], Masao Odaka[2], Akihiro Hattori[2], Kenji Yasuda[1,2] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Org. Univ. Res. Initiatives, Waseda Univ.*)

We have developed a method to fabricate the flexible three-dimensional capillary microtunnels to observe the behavior of endothelial cells in the typical inner diameter sizes of capillary blood vessels. Capillary structures in gelatin-gel were formed by melting a portion of gelatin-gel due to spot heating by a permeable 1064-nm infrared laser irradiated to the μm-sized absorption tip on the microneedle. Endothelial cells migrated and spread two-dimensionally as monolayer sheet on the inner surfaces even the diameter was 24 μm, which is similar to the cell size, instead of filling the capillary inside. We also found the velocity of leading edge of endothelial cell sheet extension in the microtunnels were inner diameter dependent and changed as inner diameter changed.

2Pos154 血管内皮細胞の二次元平面構造内における単一細胞の運動特性の観察
Behavior of single vascular endothelial cells in 2D structures

Hiromichi Hashimoto[1], Yuki Yamanaka[1], Ryuji Takano[2], Kento Iida[1], Masao Odaka[3], Kenji Matsuura[3], Akihiro Hattori[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

For understanding the mechanism of angiogenesis, we have developed methods to observe morphological index and dynamics of endothelial cells (MS-1) at single cell level. In aligned Matrigel on substrate by microfabrication, MS-1 formed sprouts between them efficiently, and tip cells size in sprouts was larger than other cells. Moreover we mixed MS-1 transfected GFP, cell shuffling was observed. In the fabricated microstructrues in an agarose thin layer which limited MS-1 migration physically, we observed single MS-1 behavior. We found that the velocity of isolated single MS-1 was faster than the MS-1 sheet extension velocity. This result suggested that MS-1 sheet velocity was not ruled by the single MS-1 velocity and some suppression factor contributed in the sheet.

2Pos155 FRET を用いた「CheYp 濃度変化」と「べん毛モーターの回転方向」の同時計測
Simultaneous measurement of flagellar motor rotation and CheYp concentration via single cell FRET

Tatsuya Yamakoshi, Yong-Suk Che, Akihiko Ishijima, Hajime Fukuoka (*Grad.Sch.Frontier.Osaka Univ.*)

In chemotaxis system in E.coli, CheYp concentration ([CheYp]) and motor-rotation are closely related, however its relation is not understood well. Therefore, we simultaneously measured the motor-rotation and [CheYp] in a single cell via FRET between CheY and CheZ. We succeeded in quantitatively analyzing the response to chemotactic stimuli; few seconds of rate constant for the change in [CheYp] was detected under physiological condition and the motor rapidly switch the rotation within the change in [CheYp]. We also detected the [CheYp] gradually increased during adaptation, while the motor resumed switching at the original frequency during the recovery of [CheYp]. We will discuss the relation intracellular signaling and chemotaxis by these quantitative parameters.

2Pos156 低浸透圧下における金魚ケラトサイト細胞シートの移動速度の上昇
Enhanced movement of fish keratocytes cell-sheet under low osmotic conditions

Naoto Ishijima, Hitoshi Tatsumi (*Human Information Systems Laboratory, Kanazawa Institute of Technology*)

Cultured gold fish keratocytes form cell-sheet. The movement of the cell-sheet under the different osmotic conditions, 25%, 50%, 75% and 100% osmolarity of control DMEM containing 10 % fetal calf serum was examined with time-lapse imaging. The movement was increased gradually in hypotonic solution within ten minutes, and stayed high level for 30 min. The maximum enhancement (approximately two-fold increase in the speed of cell migration) was observed in the 25% osmolarity condition. The dynamic contact formation and detachment from the substrate of individual cells in the cell-sheet under the low osmotic condition were examined with reflection interference contrast microscopy. The rate of contact formation was increased under low osmotic conditions.

2Pos157 A multi-omic approach to predict gene expression and metabolic functions from label-free spectral imaging of living cells

Arno Germond, Vipin Kumar, Tomonobu M. Watanabe (*RIKEN BDR*)

Current methods do not allow to estimate or predict the gene expression and specific metabolic functions directly from living cells. We hypothesized that the spectral signatures of molecules obtained by Raman spectroscopy are linked to the gene expression through the complex metabolic/proteomic network of the cell. We developed a unique approach to predict, in an accurate manner, the gene expression of thousands of living cells observed by microscopic spectral imaging, without any labeling nor destructive treatment. Our multi-omic approach enables the prediction of the full transcriptome of living cells with high accuracy and highlight metabolic functions for each cell lines. Escherichia coli strains exhibiting antibiotic resistance are used as a case study.

2Pos158 Conduction Pathway Analysis of Line-Networked Cardiomyocytes By using Multi-Electrode Array System

Tetsuro Yoshida, Tomoyuki Kaneko (*LaRC, FB, Grad. Sci.&Eng., Hosei Univ.*)

Multi-Electrode Array (MEA) system is recently expected as a new tool for cardiotoxicity test which is necessary to produce new medicines. In this study, we would like to propose conduction velocity as a new index for the test and aimed to establish the protocol to measure it. We constructed the different width of line-networked cardiomyocytes on the MEA system with agarose micro-chamber(AMC) to regulate the conducting direction, and we analyzed the conduction pathway. As a result, all width of the cardiomyocytes had generally conducted in one direction otherwise the wide one had a large fluctuation of cardiomyocytes. We are applying this method to cardiotoxicity test to measure the side effect of false negative drugs which was difficult to find with current indexes.

2Pos159 原子間力顕微鏡を用いた悪性度が異なるがん細胞の細胞間接着強度の比較
Comparison of intercellular adhesion strengths of cancer cells having different malignancies studied by atomic force microscopy

Kenta Ishibashi[1,2], Tomoko Okada[1], Chikashi Nakamura[1,2,3], Hyonchol Kim[1,2,3] ([1]*Biomed. Res. Inst., AIST*, [2]*Grad. Sch. Eng., Tokyo Univ. Agric. Technol.*, [3]*PhotoBio-OIL, AIST-Osaka Univ.*)

Intercellular adhesion strengths change dynamically in the process of tumor metastasis, however, the detailed mechanism is not well known. To understand it, the strengths of cancer cells having different malignancies were measured by using cup-attached AFM chip. Firstly, the strengths between homogeneous two cancer cells were measured and in results, the strength between highly metastatic cancer cells was larger than that between low metastatic cells. Additionally, the strengths of those two cancer cells against a vascular endothelial cell were measured, and the strength of highly metastatic cell was larger. These results indicate a possibility that highly metastatic cancer cells adhere to neighbor cells strongly in specific stages of tumor metastasis.

2Pos160 Analysis of Physical Effect on Macrophage with Agarose Microchamber

Tomoyuki Irisawa, Nami Morizino, Tomohiro Saito, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

Macrophages have an important role in inflammation and repair. A lot of previous studies have shown that macrophage functions are regulated by chemical and biologic factors. However, little is known about effects of physical setting where macrophages live. Therefore, we saw how physical factor have effects on the activity of the macrophage. The use of agarose microchamber(AMC), patterning of agarose, allowed us to observe macrophages at single cell level. Making a various width of AMC and putting one macrophage on it respectively, we examined the movement of macrophage on the AMC. As a result, we could observed the activated macrophage on the AMC in a narrow width. This experience could lead to elucidation of physical effects of the tissue structure on macrophages.

2Pos161 擬似心臓モデルとしての直線状心筋・線維芽細胞ネットワークの再構成および電気生理学性質の評価
Reconstruction of cardiac tissue-like cell structure and electrophysiological property evaluation

Koki Fujii, Tomoyuki Kaneko (*Grad. Sch. FB LaRC, Hosei Univ.*)

Cardiac tissue in vivo has a structure that cardiomyocytes (CMs) and fibroblasts (Fibs) are regularly oriented. While, cardiac cell sheet obtained by dispersed culture has scattered sarcomere structure and random contacts of CMs and Fibs. Therefore, there is an issue that they show the different functions such as contractile force and electrophysiological property. In this study, we obtained cardiac tissue-like structure on a substrate with agarose microchamber, where Fib network is localized between two Linear-CM communities and mediates their signal conduction. The CM-Fib-CM network resembling cardiac tissue in vivo was reconstructed using micro patterning of agarose gel and conducted membrane potential measurement using multi electrode array system.

2Pos162 多電極電位システムを用いた赤外線レーザー照射による心臓組織片の拍動変化
Beating rate change of heart tissue piece by infrared laser irradiation using Multi Electrode Array system

Koji Emura, Tomoyuki Kaneko (*LaRC, FB, Grad.Sci&Eng, Hosei Univ.*)

In recent years, it was found that cells exhibit various responses to external stimulation. It has been attempted to control the heart beat by noninvasive approach. In this study, a heart tissue piece from a chick embryo was exposed to an infrared laser. The tissue piece was measured the beating rate change due to laser irradiation on the electrode using Multi Electrode Array system. We changed the distance between the laser irradiation position and the heart tissue piece. Beating rate of heart tissue got faster under the condition of laser irradiation. As the laser was irradiated, the changes of beating rate was larger. There was smaller changes to irradiate the laser at father position. The heart tissue piece with accelerated beating rate paced at a constant rhythm.

2Pos163 ニワトリ胚由来心筋細胞を低温から温度を上げた際の拍動周期の変化
Beating rate change of chick embryonic cardiomyocytes heated up from low temperature

Kohei Oyama, Wei Wang, Tomoyuki Kaneko (*LaRC, FB, HOSEI unv.*)

Beating rate of cardiomyocytes was known to change depending on temperature. However, there are a little research related to beating rate change regarding rapidly temperature change in a short time. Therefore, we attempted to investigate that beating rate change as temperature of the medium turns from low temperature to body temperature. In this study we used cardiomyocytes isolated from 7-day-chick embryos and measured beating rate change under the condition of temperature change using Multi-Electrode Array system. Moreover, the change of beating rate was evaluated in terms of inter-spike interval every 1 minute during measurement. As a result, beating rate got faster as temperature was raised, and fluctuation like arrhythmia was observed in the moderate temperature.

2Pos164 心臓組織片の薬剤感受性におけるサイズ依存性の分析
Analysis of sensitivity for drugs depending size of cardiac tissue

Ryohei Kobayashi, Koji Emura, Tomoyuki Kaneko (*LaRC,FB,Hosei Univ.*)

Arrhythmia is one of the lethal diseases and often induced by drugs. For the safety assessment at an early stage of drug development, Multi Electrode Array (MEA) system is expected to detect the risk of arrhythmia. In this study, we aimed to reveal the correlation between heart tissue size and drug sensitivity, and to search for optimal conditions for cardiotoxicity test. We measured the extracellular potential and size of cardiac tissue pieces on MEA electrode, and calculated Field Potential Duration, the time from the start to the end of the beat and Short Term Variability, Pulsating temporal fluctuation. In addition, E-4031, a potassium channel blocker, is used to examine the size-dependent drug sensitivity of each piece of cardiac tissue.

2Pos165 ハイドロゲル上でのマスト細胞の脱顆粒抑制機構の研究
Inhibition of degranulation in mast cells attached to a hydrogel through defective microtubule tracts

Tadahide Furuno, Atsushi Shiki, Satoru Yokawa, Yoshikazu Inoh (*Sch. Pharm., Aichi Gakuin Univ.*)

Mast cells adhere to extracellular matrix with broad stiffness in the body. Here we compared cellular responses in mast cells on glass-base dishes with and without a hydrogel. In cells on hydrogel-coated dishes, an antigen-induced calcium response was suppressed slightly, whereas their subsequent degranulation was largely inhibited. Vinculin was distributed in a dot-like manner in resting cells on non-coated dishes but not on hydrogel-coated dishes. Microtubule reorganization and acetylation were also suppressed in activated cells adherent to the hydrogel. These suggest that adhesion to a hydrogel led to failure of composition of functional focal adhesions and microtubule tracts, which resulted in suppression of mast cell degranulation following antigen stimulation.

2Pos166 局所麻酔薬によるラフト様/非ラフト様相分離の解消とその機序
Mechanism of the local anesthetics-induced perturbation of raft-like ordered membrane domains

Masanao Kinoshita, Takeshi Chitose, Nobuaki Matsumori (*Kyushu University*)

Lipid rafts; ordered membrane domains, are thought to be possible targets of the local anesthetics (LAs) since anesthetically relevant sodium channels are recruited in the rafts. However, influence of LAs on the raft membranes remains unknown. Thus, we examined impact of representative LAs; dibuaine (Dib) and tetracaine (Tet) and lidocaine (Lid), on the raft-like ordered (Lo) and disordered phase separation. As a result, Dib the most effectively perturbs the Lo membrane and eliminates the phase separation, followed by Tet, while Lid dose the least. This ranking for the membrane perturbation; Dib > Tet > Lid, is consistent with their anesthetics potency。These results indicate the mechanistic link between disruption of lipid rafts and anesthetic action.

2Pos167 Comparative study on organizations of human stratum corneum intercellular lipids collected from various body sites

Kenta Moriwaki, Hiromitsu Nakazawa, Satoru Kato (*Grad. Sch. Sci & Tech., Univ. Kwansei Gakuin*)

We examined the organizations of human stratum corneum (SC) intercellular lipids from various body sites to clarify the influence of environmental stimulus on the skin barrier function. The transepidermal water loss (TEWL), a representative indicator of skin barrier function, as well as the intercellular lipid organization is known to vary depending on body sites. However, no extensive study on their correlation has carried out. In this study, we collected SC samples non-invasively from as many body sites as possible and analyzed the intercellular lipid organization by the low-flux electron diffraction method developed in our laboratory. We discuss about relationship of TEWL to the obtained structural data and corneocyte morphology examined in previous study.

2Pos168* 回折 X 線ブリンキング法を用いた生細胞上の GPCR 分子内部運動の決定
Determining Intramolecular Motion of GPCRs on Live Cells using Diffracted X-ray Blinking Technique

Masaki Ishihara[1,2], Shoko Fujimiura[2], Kohei Ichiyanagi[3,4], Shunsuke Nozawa[3], Shinichi Adachi[3], Ryo Fukaya[3], Masahiro Kuramochi[1,2], Kazuhiro Mio[2], Yuji Sasaki[1,2] ([1]*Grad Sch. of Fron. Sci., Univ. of Tokyo*, [2]*Univ. of Tokyo - AIST OIL*, [3]*KEK*, [4]*Jichi Med. Univ*)

G protein-coupled receptors (GPCRs) transmit extracellular signals inside the cells via activation of G proteins. GPCRs are involved in a wide variety of physiological functions. However, little has been known about the internal molecular dynamics. In this study, we measured the dynamic intramolecular motion of GPCR expressed on the cell surface at single molecular level using Diffracted X-ray Blinking (DXB) method. DXB is an X-ray based single-molecule observation technique. As a result, we found that in the event of ligand binding, the movement of the serotonin receptor (5-HT2A) and the fluctuating motion velocity of the cell membrane is intensified.

2Pos169* 大腸菌の封入密度に依存したリポソームの形態変化
Morphological changes of liposomes depending on density of encapsulated E. coli

Mai Hayakawa, Hazuki Terajima, Masahito Hayashi, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

Liposomes have the property to change their shapes by physical force. In this study, we encapsulated E. coli into liposomes as a force supplier and using emulsion transfer method. Although E. coli sometimes pushed the membrane, liposomes kept their shapes stable at low density of E. coli. Whereas, at high density of E. coli, liposomes deformed their shapes. Additionally, high density liposomes were repeatedly and rapidly changing their shapes. These results implied that liposomes showed morphological changes under the condition of the total force of encapsulated E. coli were concentrated at same membrane compartment. Our findings will be used as a new approach for generating morphological changes of liposomes.

2Pos170* 抗菌ペプチド・ラクトフェリシン B が誘起する細胞膜や脂質膜の急速な膜透過には膜電位が重要な役割を果たす
Membrane potential is vital for rapid permeabilization of plasma membranes and lipid bilayers by the antimicrobial peptide lactoferricin B

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Antimicrobial activity of lactoferricin B (LfcinB) is suggested to be due to damage of bacterial plasma membrane. Here we examined the effect of membrane potential, Δφ on LfcinB-induced damage (1). LfcinB induced rapid calcein leakage from single *E. coli* cells and spheroplasts, and a protonophore suppressed this leakage. LfcinB stochastically induced local rupture in single GUVs of *E. coli* lipid, causing rapid leakage of AF647; however higher LfcinB concentrations were required. To identify this reason, we examined the effect of Δφ on LfcinB-induced permeabilization in GUVs, finding that the rate of LfcinB-induced local rupture in GUVs increased with |Δφ|. Based on these results, we discuss the mechanism of LfcinB's antimicrobial activity.
(1) J. Biol, Chem. in press.

2Pos171* 細胞透過ペプチド・トランスポータン 10 の単一ベシクル内腔への侵入に対する膜電位の効果とそのメカニズム
Effect of membrane potential on the entry of cell-penetrating peptide transportan10 into the lumen of single vesicles and its mechanism

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Cell-penetrating peptide, transportan10 (TP10) has the permeability of plasma membrane, but its mechanistic action and related factors are still unclear. To reveal the mechanism of cell penetration, we examined the effect of membrane potential, φ_m, on the entry of fluorescent probe-labeled TP10 (CF-TP10) into vesicle lumen using single GUV method (1,2). K^+ concentration gradient was used to induce φ_m across the GUV membrane, which was confirmed by the potential sensitive-fluorescence probe. The rim intensity and the rate of entry of CF-TP10 into the GUV lumen without pore formation, increased with $|\varphi_m|$. Based on the experimental data, we discuss the mechanism of entry of CF-TP10 into single vesicle.
(1) Biochemistry, 53, 586, 2014 (2) Chem. Phys. Lipids. 212,120, 2018

2Pos172 膜タンパク質の分子内部動態解析技術の開発
Understanding intramolecular dynamics of membrane proteins using X-ray based analysis techniques

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Membrane proteins transmit extracellular signals inside cells in response to chemical and physical stimuli by changing their conformations. There are many approaches available now to solve the structures of proteins, but quite few techniques can be used to understand the real motion of protein dynamics. We adopted the Diffracted X-ray Tracking (DXT) technique. In DXT, individual protein was labelled with a gold nanocrystal and its intramolecular movement was investigated by tracking the diffraction spot. Recently, we also developed Diffracted X-ray Blinking (DXB) method. DXB uses monochromatic synchrotron radiation and laboratory X-ray source, and can suppress damage to samples, so that we could detect intramolecular motion of membrane proteins even in the living cells.

2Pos173 リン脂質とアクチンとビーズの互いに結合しないもの同士によるリポソームの形態形成
Liposome morphogenesis by phospholipid, actin filament and polystyrene bead that are not bound to each other

Ryota Kojima[1], Tomo Shibuya[2], Yutaka Sumino[2], Shunsuke Tanaka[1], Masahito Hayashi[1], **Kingo Takiguchi**[1] ([1]*Department of Biological Science, Graduate School of Science, Nagoya University*, [2]*Department of Applied Physics, Tokyo University of Science*)

Giant liposomes encapsulating F-actin have been constructed to further understand the mechanisms of cell morphogenesis and movement. Recently we showed that if the F-actin concentration in liposomes was comparable to that of cytoplasm of living cells, the liposomes could be deformed into spindle shapes by encapsulating only F-actins, even without myosins or crosslinkers. The results indicate that nematic liquid crystal formation of F-actins is important for the deformation. In this study, polystyrene beads are co-encapsulated as obstacles against the nematic phase formation. As a result, deformations other than spindle shape are observed, indicating that beads that have no unique interaction with actin can cooperate with F-actins to affect the morphology of liposomes.

2Pos174 タンパク質吸着による脂質膜の基板からの剥離
Membrane detachment from substrate induced by protein adhesion

Hiroshi Noguchi (*ISSP, Univ. Tokyo*)

We studied the detachment dynamics of a fluid membrane from a flat substrate using meshless membrane simulations. A membrane bound on a solid substrate is widely used to investigate the interaction between membrane and proteins. Recently, Boye et al. reported that lipid membranes can be detached from the substrate by the adhesion of protein annexin and the membrane form a roll structure (Nature Comm. 8, 1623). We consider the protein induces an isotropic spontaneous curvature and investigate how the membrane is detached from the substrate.

2Pos175 エピガロカテキンガレートが誘起する GUV の破裂のメカニズム
Mechanism of the burst of giant unilamellar vesicles induced by epigallocatechin gallate

Yukihiro Tamba[1], Mika Terada[1], Naoya Sugita[1], Masahito Yamazaki[2] ([1]*Natl Inst Tech, Suzuka Coll*, [2]*Shizuoka Univ*)

We investigated the process of burst of a giant unilamellar vesicles (GUVs) induced by epigallocatechin gallate (EGCg) and succeeded in observing the evolution of a pore in the GUV membrane with 3 ms time resolution. In this report, to clarify the mechanism of EGCg-induced bursting of GUVs, we examined the fractional change in area of single DOPC-GUV membranes using micropipette aspiration method which induced by the addition of EGCg. The interaction of EGCg with the tensed GUV expanded the area of its membrane. On the other hand, the interaction of EGCg with relatively relaxed GUV initially expanded and then reduced the area of the GUV. Based on the above results, we discuss the mechanism at the beginning of EGCg-induced bursting of vesicles.

2Pos176 平面型人工脂質二分子膜への昆虫細胞由来出芽ウイルスの融合観察
Observation of fusion between baculovirus budded virus envelopes and artificial planar bilayer lipid membrane

Azusa Oshima[1], Nahoko Kasai[1], Hiroshi Nakashima[1], Kanta Tsumoto[2], Koji Sumitomo[3] ([1]*NTT Basic Res. Labs.*, [2]*Mie Univ.*, [3]*Univ. Hyogo*)

Artificial planar bilayer lipid membranes (BLMs) are simple models of cellular systems under physically and chemically controlled conditions. In this study, we examined fusion of baculovirus-budded viruses (BVs) into artificial planer BLM on Si microwell substrate by fluorescent microscopy aiming for membrane protein reconstitution. Depending on the pH of the solution, BVs fused with the BLMs, which was enhanced at lower pH. The fluorescence labelled to the membrane proteins was also observed in the freestanding part of the BLMs as well as the supported part. Fusion of BVs will be a promising method to reconstitute membrane proteins to artificial free-standing BLM for development of bio-devices, where we can examine membrane protein activity.

2Pos177 赤外レーザー照射によるリポソームの形態変化
Morphological changes of liposomes by infrared laser irradiation

Tomoyuki Kaneko, Akira Oguri, Shunsuke Shiomi, Mai Hayakawa, Masahito Hayashi (*LaRC, FB, Hosei Univ.*)

Liposomes are well known to be changed the morphology by internal or external force (e.g., microtubule polymerization or osmotic pressure). To observe the morphological changes of liposomes by a noncontact force, we tried to irradiate the infrared (wavelength: 1480 nm) continuous-wave fiber laser to liposomes made by inverted emulsion method. As a result, the membrane of liposomes was observed to be fluctuated after infrared laser irradiation to liposomes. It was suggested that liposomes might be supplied with energy such as heat and flow by infrared laser. We will attempt to observe the morphological changes of liposomes encapsulated tubulin or bacteria by infrared laser irradiation.

2Pos178 抗菌ペプチド・マガイニン 2 が誘起するポア形成に対する膜電位の効果
Effect of membrane potential on antimicrobial peptide magainin 2 (mag)-induced pore formation in lipid bilayers

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Recent study suggested that membrane potential, φ_m, plays a vital role in the damage of plasma membrane by antimicrobial peptides (AMPs). However, the mechanism of the role of φ_m in AMP's activity is not clear. Here, we examined the effect of φ_m on mag-induced pore formation in single PG/PC-GUVs, because mag is an AMP whose action of mode is well characterized (1). Various membrane potentials in GUVs were generated by K^+ concentration gradient. The rate constant of mag-induced pore formation increased with increasing $|\varphi_m|$. Moreover, mag-induced fractional area change of GUV membrane (2) increased with increasing $|\varphi_m|$. Based on the experimental results, we discuss the effect of φ_m on mag-induced pore formation.
(1) Langmuir, 34, 3349, 2018 (2) Langmuir, 31, 3391, 2015

2Pos179 蛍光プローブでラベルされていない細胞透過ペプチド・トランスポータン 10 と単一巨大リポソームとの相互作用
Interaction of non-fluorescent probe-labelled cell-penetrating peptide transportan 10 with single giant unilamellar vesicles (GUVs)

Madhabi Shuma[1], Md. Mizanur Moghal[1], Masahito Yamazaki[1,2,3] ([1]*Grad. Sch. Sci. Tech., Shizuoka Univ.*, [2]*Res. Inst. Ele., Shizuoka Univ.*, [3]*Grad. Sch. Sci., Shizuoka Univ.*)

To investigate the entry of cell-penetrating peptide (CPPs) into vesicle lumen and cytosol, fluorescent probe-labelled CPPs have been used (1). However, the labelling may affect the interaction of CPPs with lipid bilayers. Here, we investigated the interactions of a non-labelled CPP, transportan 10 (TP 10) with single PG/PC-GUVs. First, we examined the flip-flop or trans-bilayer movement of NBD-PG during the interaction of TP 10 with single GUVs using a new method (2). The rate of flip-flop greatly increased with TP 10 concentration before pore formation. We discuss the implications of this result as well as other experimental results of the interaction with single GUVs.
(1) Appl. Microbiol. Biotechnol. 102, 3879, 2018. (2) J. Chem. Phys. 148, 245101, 2018.

2Pos180 浸透圧により DOPG/DOPC-GUV に誘起される膜張力の評価
Estimation of Membrane Tension of DOPG/DOPC-GUVs Induced by Osmotic Pressure

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Osmotic pressure, Π, induces lateral tension in plasma membrane and lipid bilayers, which plays important roles in function of membranes. Recently, membrane tension, σ_{osm}, in DOPC-GUVs induced by Π has been quantitatively estimated using a new method (1). Here we examined effect of Π on negatively charged DOPG/DOPC-GUVs. First, we investigated the effect of Π on the rate constant, k_p, of constant tension-induced rupture of GUVs, and based on its analysis the values of σ_{osm} under various values of Π were estimated. The values of σ_{osm} in DOPG/DOPC-GUVs were smaller than those in DOPC-GUVs under the same Π. We also investigated Π-induced pore formation in DOPG/DOPC-GUVs. Based on these results, we discuss the effect of Π on DOPG/DOPC-GUVs.
(1) Biophys. J, 111, 2190, 2016

2Pos181 抗菌性オリゴ糖が誘発する脂質二分子膜の多層化
An antibacterial oligosaccharide makes lipid bilayer multi-layered

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A cyclodextrin (CD) derivative, whose one end face of cylinder is modified by alkylamino group, induces damage of bacterial membranes and kill bacteria. However, morphological change of membrane during damaging is not cleared yet. Here, we observed antibacterial CD derivative-induced morphological change of lipid bilayer by AFM. We used DOPG bilayer for model of negatively charged bacterial membrane. Addition of the antibacterial CD derivative made the bilayer multi-layered within 30 minutes, though cyclodextrin didn't. The result suggests that positively charged and hydrophobic moiety of the derivative facilitates distribution to negatively-charged surface and hydrophobic core of DOPG bilayer, increasing bilayer-bilayer interaction.

2Pos182 Leaflet-specific lipid diffusions in supported lipid bilayers

Takuhiro Otosu, Shoichi Yamaguchi (*Saitama Univ.*)

A lipid bilayer supported on a solid substrate (SLB) is now widely used as a model biomembrane due to its high stability and the fluidity comparable to a free-standing bilayer. However, the effect of a solid support on the lipid diffusion in two layers (leaflets) of the SLB remains fully elusive. In this regard, we previously demonstrated that two-dimensional fluorescence lifetime correlation spectroscopy (2D FLCS) is a powerful tool to quantitatively examine the lipid diffusion in each leaflet of SLBs. In this presentation, we discuss the results of leaflet-specific lipid diffusions performed on various SLBs.

2Pos183 インクジェット塗布を用いたパターン化人工生体膜の開発
Inkjet-printed and dried lipid membrane arrays for the biophysical studies and biosensing applications

Yasushi Tanimoto[1], Misato Yamada[2], Fumio Hayashi[3], Kenichi Morigaki[1,2] ([1]*Biosignal, Kobe Univ.*, [2]*Grad. Sch. Agr., Univ. Kobe*, [3]*Grad. Sch. Sci., Univ. Kobe*)

Biological membranes containing membrane proteins are a natural biosensing unit, whose functions can be exploited in the biophysical studies and biosensing applications. For realizing membrane-based biosensors, we developed a methodology to generate arrays of lipid bilayers on a solid substrate by printing lipids in an organic solvent and drying the solvent. Upon hydration, printed lipid patches spontaneously swell and spread to form supported bilayers. We demonstrate lipid-protein interactions such as biotin-streptavidin binding and reconstitution of integral membrane protein, rhodopsin in the addressable arrays. The printed and dried lipids provide unique opportunities to be stored for a prolonged period and applied onsite by the rapid hydration.

2Pos184 Multiscale molecular dynamics simulations of F-BAR protein Pacsin1: Assembly and curvature preference on lipid membrane

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F-BAR (F-Bin/Amphiphysin/Rvs) domain proteins are membrane-associated proteins that are involved in membrane remodeling via binding to the membrane. Notably, Pacsin1 has an ability to transform membrane into different morphologies: striated tubes, featureless wide and thin tubes, and pearling vesicles. Molecular mechanism of this interesting ability remains elusive. We perform all-atom and coarse-grained MD simulations to investigate assembly and curvature sensing of Pacsin1 on the membrane. The regularly assembled Pacsin1 dimers bend the tensionless membrane. With the single dimer, however, it senses the membrane curvature and binds to the top of the buckled membrane with a preferred angle. These results provide molecular insights into how Pacsin1 remodels membrane.

2Pos185 高速 AFM によるハブ毒液由来のホスホリパーゼ A_2 によって引き起こされる膜分解の動態観察
Membrane degradation dynamics by phospholipase A_2 from snake venom observed by high-speed AFM

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Phospholipase A_2 (PLA_2) is a ubiquitous enzyme that hydrolyzes phospholipids. Habu snake venom contains multiple PLA_2 isozymes, of which PLA2 and BPII are major components. Cytotoxicity of BPll is much higher than that of PLA2, although lecithin degradation activity of BPII is very lower. Here, we analyzed the membrane degradation process by high-speed AFM. The area of lipid bilayers substantially decreased upon addition of PLA2, in comparison to the addition of BPII, with which only a little degraded. BPII may have other functions than lipid hydrolysis on cytotoxicity. For PLA2, the degradation speed at the edge of bilayer was much faster than at the continuous surface of the bilayer, and the existence of PE on the membrane facilitated the reaction.

2Pos186 グラフニューラルネットワークを用いた運動想起時脳波分類
Classification of Motor Imagery Using Graph Neural Networks

Ryo Nakajima[1], Hideo Mukai[2] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci. & Tech., Meiji Univ.*)

We analyzed motor imagery electroencephalography (EEG) by Graph Neural Networks (GNN). Because it is known that EEG has correlation between different brain regions, we regard EEG data as signals on graph. To extract intended motor signals from EEG, we used GNN, which is an effective framework for representation learning and prediction for graph structural data. The EEG signals of each electrode were divided into multiple frequency bands. Then we merged graphs builded for the same frequency band to obtain brain graph representation of EEG. To evaluate the effectiveness of GNN models, we used motor imagery data of left and right limbs. A few of these models showed improvement of accuracy compared to other deep learning methods.

2Pos187 運動想起時脳波の時間周波数解析とニューラルネットワークによる識別
Time-Frequency and neural network analysis for classification motor imagery EEG

Azumi Ohno[1], Hideo Mukai[1,2] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci. & Tech., Meiji Univ.*)

Motor imagery classification task based on electroencephalogram (EEG) is an important issue in brain-computer interface. However there still remains room for improvements in terms of classification success rate. To address this problem, we tried to improve accuracy of classification targeting on feature extraction. We focused to extract ERD/ERS responses in the motor imagery EEG, because these signals have features corresponding to movement intention. To obtain the features of EEG, we used Short-Time Fourier Transform (STFT) and Wavelet Transform (WT). After applying these methods we classified motor imagery signals by convolutional neural network. Our current results suggest that STFT performs better than WT without hyperparameter tuning.

2Pos188 深層畳み込みニューラルネットワークを用いたマウスの社会的行動の自動検出
Automated detection of social behavior in mice using deep convolutional neural network

Hideo Mukai[1,2], **Kenji Takemoto**[1] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech., Meiji Univ.*, [2]*Dept. Comp. Sci., Sch. Sci. & Tech., Meiji Univ.*)

We analyzed the social behavior of mice from recorded images by machine learning using convolutional neural network(CNN). Making remarkable achievements in computer vision, CNN performs not only accurate detection of actions but classification based on unified criteria, which are rather difficult by simple inspection. We adopted the semantic segmentation to assign a class label to each pixel in the images. To investigate emotional response, we placed mice in the same cage after a mouse observed a conspecific receiving foot shock. These animals showed increased empathic behavior such as affiliative contacts and freezing responses compared to the case without any shock. It is suggested mice have the capacity to recognize conspecific's states suffering from distress.

2Pos189 仮想空間を用いた EEG による運動想起フィードバック訓練システムの構築
EEG-Based motor imagery feedback training system on VR environment

Hideo Mukai[1,2], **Kazuki Kobayashi**[1] ([1]*Comp. Sci. Prog., Grad. Sch. Sci. & Tech.,Meiji Univ.*, [2]*Dept. Comp.Sci., Sch. Sci & Tech., Meiji Univ*)

In this study we developed a sensory feedback system with brain-computer interface(BCI) for rehabilitation. When motor disorder is caused by a stroke, etc., motor learning together with sensory feedback fascilitates recovery for damaged function. Therefore, we tried to build a training system of electroencephalogram(EEG) signal in motor imagery using virtual reality(VR). First, we designed the avatar of arms controllable by EEG in VR engine(Unreal Engine 4). Next, we implemented a subject training BCI-system that gives feedback corresponding to exercise intention, then examined its training effect. The results showed that motor imagery feedback training on VR environment were effective even in a short period of time.

2Pos190 線虫 C.elegans の学習行動を制御する神経回路のシナプス可塑性
Mechanisms of Synaptic Plasticity in a Neural Circuit that Regulates Memory Dependent Behavior in C. elegans

Llian Mabardi, Hirofumi Kunitomo, Hirofumi Sato, Yu Toyoshima, Yuichi Iino (*Tokyo University School fo Science Department of Biology*)

To successfully navigate their environment animals need to receive, encode and store sensory input, and retrieve these memories to make decisions. C. elegans sense ambient salt concentrations using the ASE sensory neuron pair. Downstream are the interneurons AIA, AIB and AIY which interpret signals from sensory neurons and then signal downstream to other interneurons and motor neurons to drive behavior. We seek to uncover the mechanisms of memory formation and learning by using calcium imaging to study plasticity of AIY, specifically focusing on changes that occur between the ASE neuron pair and AIY that occur after experiencing differing salt concentrations in the presence of food.

2Pos191 海馬で合成される男性・女性ホルモンやストレスホルモンによる記憶シナプスの早い non-genomic な制御
Rapid non-genomic modulation of synapses by hippocampus-synthesized androgen, estrogen and stress steroid

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Neurosteroids (sex steroids and stress steroids) are synthesized in the hippocampus, center for learning and memory. Rapid action of neurosteroids has been extensively studied in the hippocampus over more than decades, and a significant progress has been achieved in clarification of essential molecular mechanisms. Dihydrotestosterone (DHT), testosterone (T), estradiol (E2) and corticosterone trigger synaptic (membrane) sex steroid receptors and glucocorticoid receptor, leading to rapid modulation of dendritic spines in hippocampal slices. In downstream, kinase-dependent signaling is involved in non-genomic modulation of dendritic spinogenesis of not only sex steroids but also stress steroids.

2Pos192 A leadership-based phase transition in a flocking model with activated and un-activated agents

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Flocking models are often used to understand collective motion. Usually a set of homogeneous agents begins in a disordered state with simple local interactions and may eventually reach a cohesive state over time. We investigate the effects of allowing agents to activate or de-activate based on simple rules, much like the interaction among single-celled organisms such as colonies of the amoeba Dictyostelium Discoideum. Our study examines the effect of intermittent leadership on the cohesion of cell colonies and highlights an analogy between flocking and cell aggregation. We show that our activation-based model exhibits a sharp transition in time from incoherent to coherent states, which is seen in amoeba aggregation but not captured by other models of amoeba colonies.

2Pos193* レチナールを結合するリジンを保存しない微生物型ロドプシンの光応答性機能獲得
Engineering microbial rhodopsin without retinal-binding lysine to gain photosensitive function

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Microbial rhodopsin is a retinal-binding membrane protein with the Schiff base linkage at a lysine on the 7th helix. About 10 % of microbial rhodopsins predicted from genome sequences, however, lack retinal-binding lysine at the corresponding position (Rh-noK), suggesting that Rh-noK has function without retinal. In this study, we succeeded in gaining function of Rh-noK by introducing mutations. Two Rh-noKs from bacteria were heterologously expressed in *Escherichia coli* and showed no color. When retinal-binding lysine was introduced, one of them gained visible color. Additional mutation of the Schiff base counterion further gained proton-pumping activity. These results suggest that the Rh-noK forms the similar structure for the function to other microbial rhodopsins.

2Pos194* T(6-4)C の同位体標識を用いた(6-4)光回復酵素の低温における DNA 修復中間体の赤外分光測定
Low-temperature FTIR study of the repair intermediates of T(6-4)C/photolyase using isotope labeling

Katsuya Maeda[1], Mai Kumagai[1], Daichi Yamada[2], Yuma Terai[3], Junpei Yamamoto[3], Hideki Kandori[1] ([1]*Nagoya Inst. Tech*, [2]*Univ. Hyogo.*, [3]*Osaka Univ.*)

(6-4) photolyase is a DNA repair enzyme that reverts UV-induced (6-4) photoproducts into normal bases. (6-4) photolyase is able to repair both T(6-4)T and T(6-4)C, where the OH and NH2 groups are bound at C5 position of 5' side, respectively. Until now, researches have been conducted mainly on T(6-4)T, and the repair mechanism of T(6-4)C is believed to be the same as T(6-4)T without detailed analysis. Here, we studied the repair mechanism of T(6-4)C using of low-temperature light-induced difference FTIR spectroscopy. We attempted to identify vibrations of the repair intermediates by isotope labeling of the transferable functional group of T(6-4)C. We will discuss the mechanism by comparing the obtained results with those of T(6-4)T.

2Pos195* シネコシスティスハロロドプシン（SyHR）のアニオン輸送における 塩基性アミノ酸の機能的役割
Functional roles of basic amino acids on the anion transport in *Synechocystis* halorhodopsin (SyHR)

Masaki Nakama[1], Keiichi Kojima[1], Marie Kurihara[1], Susumu Yoshizawa[2], Yuki Sudo[1] ([1]*Grad. Sch. of Med. Dent.& Pharm. Sci. Okayama Univ.*, [2]*AORI, UTokyo*)

Microbial rhodopsins act as light-driven ion transporters, while the substrate ions have been confined to only monovalent ions such as H^+ and Cl^-. Recently, we identified a novel microbial rhodopsin from the cyanobacterium *Synechocystis* sp. PCC 7509 [JACS, 2017]. The protein named SyHR transports not only Cl^-, but also a polyatomic divalent ion, SO_4^{2-}. To understand its anion transport mechanism, we performed alanine-scanning mutagenesis on twelve basic amino acids on the transmembrane region. Among the mutants, anion transport activities showed a wide variety. Then, we analyzed the photochemical properties of the mutants to estimate the mutation effects. Based on the results, we propose a model for anion transport mechanism of SyHR.

2Pos196* Molecular characterization of heliorhodopsin from marine giant virus light-dependently infecting to *Emiliania huxleyi*

Ritsu Mizutori[1], Masae Konno[1,2], Keiichi Inoue[1,3], Oded Beja[4], Hideki Kandori[1,2] ([1]*Grad. Sch. Eng., NIT*, [2]*OBTRC, NIT*, [3]*ISSP, Univ. Tokyo*, [4]*Technion-Israel Inst. Tech.*)

New heliorhodopsins (HeRs) were found from giant viruses infecting *Emiliania huxleyi* (*Eh*Vs). They are categorized into three phylogenic groups. Here we studied photocycles and retinal isomeric compositions of three types of *Eh*V-HeR from each phylogenic group. The first group showed similar molecular properties to other bacterial HeRs showing long-lived O intermediate. The retinal composition was also similar to bacterial HeR in which all-*trans* retinal was the major isomer in the dark. The second group showed blue-shifted absorption and a long M intermediate accumulation. The third group has a characteristic C-terminal tail containing many prolines. We will present molecular properties and characteristics of the three groups in more detail.

2Pos197* 分光学的手法による霊長類青感受性視物質の光反応中間体解析
Photochemical reactions of a primate blue-sensitive pigment by spectroscopic study

Shunpei Hanai[1], Kota Katayama[1], Takuma Sasaki[1], Hiroo Imai[2], Hideki Kandori[1] ([1]*Nagoya Institute of Technology*, [2]*Primate Research Institute, Kyoto University*)

Color visual pigments contain a common 11-cis-retinal as the chromophore. Light absorption by the retinal causes cis-trans isomerization, followed by conformational changes of the protein moiety. However, molecular mechanisms of structural changes underlying signal transduction of color pigments remain unclear. Here, focusing on the blue sensitive pigment (MB), we identified all intermediates states formed by thermal relaxation from the primary Batho state using low-temperature UV-visible spectroscopy. Unlike other pigments, all intermediates except for Meta-II accumulated at specific temperature are converted into the initial state by light, which is highly advantageous for the structural analysis by FTIR spectroscopy.

2Pos198 (2SFA-6) 微生物型ロドプシンに基づく光遺伝学ツールの探索と開発
(2SFA-6) Exploration and development of microbial rhodopsin-based optogenetic tools

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Microbial rhodopsins are photoreceptive membrane proteins containing retinal as a chromophore. Genomic advances have revealed that they are widely distributed in all three biological domains with a wide variety of biological functions (e.g. ion transporters and light sensors). Recently, they serve as fundamental tools of optogenetics, a technology to regulate biological phenomena with light. Towards development of optogenetic tools, we are investigating them with bottom-up strategies as follows; (i) we explored novel rhodopsins from nature and characterized their functions and properties. (ii) we rationally modified their properties. (iii) we applied them to optogenetic tools. In this symposium, we will discuss our recent progress and future prospects.

2Pos199 ロドプシンクラスター上におけるトランスデューシンの動的過程の高速 AFM 観察
High-speed AFM observation of the dynamic process of transducin on rhodopsin cluster

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Rhodopsin (Rh) molecules form supramolecular structures such as dimers and clusters. Recent structural and simulation studies have suggested that G-protein transducin (Gt) can be efficiently activated by sliding on Rh dimer rows. However, the interaction between Rh cluster and Gt on photo-transduction process has never been directly observed. In order to clarify the functional role of Rh cluster, we observed the interaction process of Gt on Rh clusters in membrane by high-speed AFM. High speed AFM visualized Rh clusters consisting of dimer rows in membrane under physiological buffer condition. AFM movies showed that Gt molecules diffused on Rh clusters under the dark. Then, we observed that some of them transiently stayed on Rh clusters upon light illumination.

2Pos200 網膜桿体細胞内円盤膜上での脂質-光受容タンパク質秩序形成の数理モデル
A mathematical model of pattern formation of lipid-photoreceptor proteins on disk membranes of retinal cells

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Phototransduction of vertebrate starts from the disk membrane in outer segment of rod cells in the retina. Disk membrane consists of raft lipid, non-raft lipid, and rhodopsin (Rh). Various imaging studies reported the clusters of aligned Rh dimer are formed around the center of disk membrane. These studies suggested the affinities between Rh and various lipids provide important contribution to such pattern formation. However, the mechanism is unclear. We constructed a mathematical model to clarify the mechanism of aligned Rh clusters formation in disk membrane. We found the phase separations of raft and non-raft lipids and Rh-raft lipid affinity form and maintain the straight aligned Rh dimer clusters.

2Pos201 新奇チャネルロドプシン Ts_Rh3 の電気生理学的解析
Electrophysiological analysis of a novel channelrhodopsin Ts_Rh3

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We here report a novel microbial rhodopsin Ts_Rh3. Ts_Rh3 consists from a 7 transmembrane domain followed by a long cytoplasmic domain about 560 amino acids, a peptidoglycan (PG) binding domain is involved. By electrophysiological measurement after expressing in mammalian cell (ND7/23 cells), Ts_Rh3 is revealed to be a cation-conducting channelrhodopsin with λmax of 450 nm which is significantly blue-shifted compared to other ChRs. Surprisingly, deletion of cytoplasmic domain dramatically slow down the photocycle and thus its channel kinetics. These results indicate the interaction between rhodopsin domain and C-terminus domain to modulate the channel kinetics. Molecular mechanism of Ts_Rh3 will be discussed based on the electrophysiological analysis.

2Pos202 リン酸化ロドプシン･アレスチン複合体は視細胞円板膜切れ込み部に集まる
Phosphorylated-rhodopsin/arrestin complex assembles to disc incisures

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Photo-bleached rhodopsin (Rh*) catalytically activates G protein transducin (Gt) to initiate phototransduction. The inactivation process of Rh* starts by its phosphorylation, and the potency of Rh* to activate Gt is fully quenched by binding of a cytosolic protein visual-arrestin (Arr) to pRh*. Yet, the recovery process of Rh, i.e. the dissociation of pRh*-Arr and the de-phosphorylation of Rh* have remained obscure. To address these issues, we explored the behavior and distribution of pRh*-Arr complex in the bullfrog disc membrane by single-molecule and semi-multimolecule fluorescence imaging. Results show that pRh*-Arr distributes mostly at the edge of the disk membrane forming the aggregates at the incisures. Mechanism of this phenomenon would be discussed.

2Pos203 ボルボックスの光驚動反応における鞭毛運動の照度依存性
Light intensity dependence of adaptive photo-response of Volvox

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Multicellular organisms respond to external force by integrating the response of each cell. To investigate the hierarchy of the response, we focus on phototaxis of a green alga Volvox. Volvox is a multicellular organism which consisted of a few thousand somatic cells having two flagella. When Volvox is exposed to light, the flagellar beating immediately stops and recovers in approximately ten seconds. The adaptive photo-response enables Volvox to swim toward the light (phototaxis). In this presentation, the light intensity dependence of adaptive photo-response and the relationship between the adaptive photo-response and phototaxis will be discussed.

2Pos204 共鳴ラマン分光法と MD + QM/MM 計算を用いたシアノバクテリオクロム発色団の脱プロトン化部位の同定
Identification of the Deprotonated Pyrrole Nitrogen of the Bilin-Based Photoreceptor by Raman Spectroscopy with MD+QM/MM Analysis

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Phytochrome and cyanobacteriochrome utilize a linear methine-bridged tetrapyrrole (bilin) to control numerous biological processes. They show a reversible photoconversion between two spectrally distinct states. We report a resonance Raman spectroscopic study on cyanobacteriochrome RcaE, which has been proposed to contain a deprotonated bilin for its green-absorbing 15Z state. The observed Raman spectra were well reproduced by a simulated structure whose bilin B ring is deprotonated, with the aid of molecular dynamics and quantum mechanics/molecular mechanics calculations. The results revealed that the deprotonation of B and C rings has the distinct effect on the overall bilin structure, which will be relevant to the biological function in phytochrome superfamily.

2Pos205 ビリベルジン結合型シアノバクテリオクロムの遠赤／橙色光変換過程での構造変化の検出
Detection of structural change during far-red/orange reversible photoconversion of biliverdin-binding cyanobacteriaochrome

Yuka Takeda, Keiji Fushimi, Rei Narikawa (*Grad. Sch. Sci., Univ. Shizuoka*)

Cyanobacteriochromes (CBCRs) are linear tetrapyrrole-binding photoreceptors to sense wide spectral range from ultraviolet to far-red. Recently, we have discovered biliverdin (BV) -binding cyanobacteriochromes showing far-red/orange reversible photoconversion. Because BV is an intrinsic chromophore in mammalian cells and absorbs far-red light that deeply penetrates into mammalian tissues, BV-based tools should be advantageous for mammalian optogenetic tools. In this study, we aimed to reveal details of the structural change upon photoconversion. The structural change was accompanied not by the transition of the oligomeric state but rather by alteration of the exposed surface of the monomeric form. We are now trying to develop optogenetic tools based on these findings.

2Pos206 赤外分光法によって明らかになった色覚視物質とロドプシンの構造ダイナミクスの違い
Different structural dynamics between cone pigments and rhodopsin revealed by FTIR spectroscopy

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Cone visual pigments are responsible for color vision, whose structural information is much less known than that of rhodopsin. Recently, we obtained the light-induced FTIR difference spectra of Lumi intermediate of primate green pigment (MG) at >100 K. From this study, a large α-helical structural change coupled with retinal relaxation was observed. Here, we extend these studies by assessing MG conformational dynamics during activation, using ATR-FTIR spectroscopy. The Meta-II spectra measured at 300 K showed further large backbone structural change from Lumi, which would be characterized by an outward tilting of TM6 like rhodopsin, whereas different hydrogen-bonding alteration originating from a key carboxylic acid were discovered between them.

2Pos207 An Anion Channelrhodopsin with a Naturally Super-Slow Photocycle

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Focusing on the amino acid which is important for the channel gating in anion channelrhodopsins (ACRs) reported to date, ACRs are divided into three subtypes, named Cys-, Thr-, and Val-types. Here we investigated the ACRs from a cryptophyte algae identified in Antractica, *Geminigera cryophila*, because this algae has the ACRs belonging to all three subtypes. We successfully expressed the Cys-, Thr-, and Val-type *Gc*ACRs and measured their photocycle kinetics. As a result, the Val-type *Gc*ACR has a significantly slow photocycle (~ 200 s) by comparing to that of the conventional Cys-type ACRs (~ 1 s). We will discuss in detail the molecular properties of the Val-type *Gc*ACR in this meeting.

2Pos208 赤外分光法により明らかとなった酵素型ロドプシンの構造的特徴
Structural features of enzyme rhodopsins revealed by infrared spectroscopy

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In recent years, a new type of microbial rhodopsin has been discovered, in which enzyme is linked to the photoreceptor protein rhodopsin. Rh-GC synthesizes cGMP, while Rh-PDE degrades cGMP and cAMP. Biochemical experiments have shown these light-dependent enzyme activities, but the reaction mechanism has not been elucidated. Here, we attempted to monitor the structural changes of the enzyme rhodopsins to grasp the light regulation mechanism using FTIR spectroscopy. Based on the results obtained, we will compare difference in the structural changes in membrane domains between Rh-GC and Rh-PDE to find a key step for activating these different enzymatic activities.

2Pos209 *Mastigocladopsis repens* halorhodopsin の Cl^-ポンプ活性における His166 の重要性
Importance of His166 for Cl^--pump activity of *Mastigocladopsis repens* halorhodopsin

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Mastigocladopsis repens halorhodopsin (MrHR) is a thirdly identified Cl^- pump rhodopsin. Compared to other Cl^- pumps, MrHR shows slower photocycle and conserves the residues essential for H^+ pumps. Thus, MrHR probably evolved from H^+ pump but has not fully developed to matured Cl^- pump. Here, we report an importance of His166 in the hydrophobic cytoplasmic (CP) channel. The pump activity was revealed to need highly hydrophilic residue at this position. In other Cl^- pumps, this residue is replaced with Thr or Phe, and their mutations did not affect the pump activities. These pumps also have hydrophobic CP channel and seem to require transient hydrations for the Cl^- transports. MrHR might not cause the hydration and thus need highly hydrophilic residue in this channel.

2Pos210 時間分解フーリエ変換赤外分光法による KR2 のナトリウムおよびリチウムイオン輸送の分子機構研究
Time-resolved FTIR spectroscopy for studying molecular mechanisms of sodium and lithium ion transportation of *Krokinobacter* rhodopsin 2

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KR2 is the first discovered sodium pump rhodopsin, which can pump sodium and lithium ions, and also protons in the absence of the former ions. In the photocycle of KR2, it is assumed that each of the cations passes through the Schiff base most likely upon the formation of the O intermediate. Here, we applied light-induced step-scan FTIR spectroscopy to study structural changes of KR2 during sodium and lithium ion transport. From global fitting analysis, we detected several intermediates in the photocycle, K, L/M, O_1, and O_2. Two O intermediates with distinct spectral feature has been successfully observed in KR2. Moreover, we detected difference in decay kinetics between lithium and sodium pump. The molecular mechanisms of cation transportation in KR2 will be discussed.

2Pos211 Rc-PYP の多量体複合体形成における K72 の役割
Functional role of a residue K72 of Rc-PYP in light dependent oligomeric complex formation process

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Rhodobacter capsulatus PYP (Rc-PYP) is a light receptor protein which binds to PYP binding protein (PBP) in a light-dependent manner. Our previous titration SAXS experiments revealed that Rc-PYP and PBP exhibit various oligomeric forms. The fact implies PYP would have two distinct binding sites against PBP, which have not been identified. A K72Q mutant showed almost identical UV-Vis absorption to that of wild-type, bur the K72Q partially lost its binding ability to PBP. Titration measurement by SAXS of K72Q against PBP revealed that K72Q mutant has less binding ability against PBP, especially in oligomeric complex formation process. This result pointed out that surface located residue K72 is included oligomeric complex interaction interface.

2Pos212 FTIR study on the localization of the excited triplet state of chlorophyll in photosystem II

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The excited triplet state of chlorophyll (^{3}Chl*) in PSII is a precursor of harmful $^1O_2$* and functions as a switch from a protective to degradative phase. Although ^{3}Chl* is known to be mainly located on Chl_{D1}, Chl molecules with which ^{3}Chl* is equilibrated remains unidentified. Here, we investigated the ^{3}Chl* localization by measuring triplet/singlet FTIR difference spectra with D1-V157H and D2-V156H mutants, in which an H-bond was introduced to the keto C=O of P_{D1} and P_{D2}, respectively. Upon D2-V156H mutation, prominent triplet/singlet keto C=O bands due to P_{D2} were observed at 1633/1680 cm^{-1}, while new shoulders due probably to P_{D1} appeared at ~1640/~1695 cm^{-1} upon D1-V157H mutation. These results indicate that ^{3}Chl* is located over Chl_{D1}, P_{D1} and P_{D2} in equilibrium.

2Pos213 緑色イオウ細菌 Chlorobaculum tepidum からの反応中心複合体標品の改良
Improved preparation of the reaction center complex from the green sulfur bacterium *Chlorobaculum tepidum*

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The reaction center (RC) of green sulfur bacteria is the homodimeric type 1 RC, and its electron transfer scheme is supposed to be analogous to that in photosystem I. We have developed a gene expression system by introducing a plasmid pDSK5191 or 5192 stably harboring the second pscAB gene cluster, and as a result, a lot of amount of the His/His-tagged homodimeric RC could easily be obtained (Azai et al., 2013). However, some subunits tended to be dissociated from the complex during purification. In this study, we improved the purification method in order to hold all subunits as stable as possible under a dim-light condition. The amplitude of the oxidized $P840^+$ by the flash was found to be much larger than before. We now tried to crystallize this improved preparation.

2Pos214 Carotenoid glycoside quenches bacteriochlorophyll *a* fluorescence in the photosynthetic reaction center complex of green sulfur bacteria

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Carotenoid is found in all the photosynthetic organisms, and used for harvesting or dissipation of light energy in photosynthetic pigment-protein complexes. Some of carotenoid are glycosylated at their one edge of the polyene backbone. However, their function has remained unknown, especially in terms of the effect of glycosylation on photosynthesis. Here, we present a photosynthetic function of 1'-hydroxy-γ-carotene glucoside ester, which is bound to the reaction center complex in the green sulfur bacterium *Chlorobaculum tepidum*. Genetic deletion of the glycosylation specifically prolonged a fluorescence lifetime of bacteriochlorophyll *a* in the reaction center complex. The glycosylation would promote dissipation of excess excitation energy in *C. tepidum*.

2Pos215 Isolation of the Rieske/cytochrome *b* complex from green sulfur bacteria and interaction of the Rieske protein with cytochrome *c*-556

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The cytochrome (cyt) *bc* complex is a membrane protein complex which functions in the electron transport chain. Generally, this complex is composed of three subunits, cyt *b*, Rieske protein, and cyt *c* (c_1 or *f*). Green sulfur bacteria contain the Rieske/cyt*b* complex which lacks cyt *c*. This complex has been supposed to have the most primitive structure and function. In this study, we examined solubilization and purification of this complex to reveal its crystal structure. Since a cofactor heme *b* was easily dissociated from cyt *b* under aerobic conditions, we are now trying to isolate it under anaerobic conditions. We also observed the interaction between the Rieske protein and cyt *c*-556 by NMR measurements and identified the interaction sites on the Rieske protein.

2Pos216 Spectroscopic characterization of a bacteriochlorophyll b-based LH1-RC complexes from thermophilic purple bacterium Blactochloris tepida

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Blactochloris (Blc.) tepida is a thermophilic purple nonsulfur bacterium which grows optimally above 40 C(up to 47C). The Light-harvesting 1 reaction center (LH1-RC) complex contains bacteriochlorophyll (BChl) b and exhibits its LH1 Qy peak at ~1006 nm, largely red-shifted from those of LH1-RCs containing BChl a. A recent structural study revealed that the LH1 of the mesophilic counterpart Blc. viridis is comprised of triple rings and the hole lacking one gamma-subunit was predicted to be a gate transporting redox-active quinones. In the present study, we investigated spectroscopically the origins of the unusually red-shifted Qy absorption, the enhanced thermal stability, and light-induced quinone reduction of Blc. tepida compared with those of Blc. viridis.

2Pos217 光感受性アデニル酸シクラーゼ OaPAC の活性制御部位の同定
Identification of the activity-regulating site in the photoactivated adenylate cyclase (OaPAC)

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The photoactivated adenylyl cyclase from the Oscillatoria acuminata (OaPAC) which produces cAMP by illumination with blue-light is an attractive tool to regulate cellular events with precise spatial and temporal control with light. However, a mechanism of regulating the adenylate cyclase activity in OaPAC has not been clear. To clarify the mechanism, we made several mutants and measured cAMP-dependent luminescence in HEK cells. Some mutants in which a specific region was mutated showed much higher luminescence than wild-type, indicating that activities of these mutants were much higher than wild-type. Therefore, it suggests that the region inhibits cAMP production and play an important role in regulating the activity of OaPAC.

2Pos218 Light-dependent structural states of OaPAC

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OaPAC is one of the photoactivated adenylyl cyclases (PAC) which produce cAMP from ATP in response to blue light. The crystal structure of OaPAC has been determined, however, the dynamic structural changes with photoactivation has not been clear. In this study, we aim to reveal structural states around the ATP binding site under dark or photo-activated conditions. Using environment-dependent fluorescent dye, TMR, we measured the fluorescence under dark or blue-light irradiated conditions and detected the structural changes as changes in the intensities. We will show the light-dependent structural states and discuss how light-dependent structural changes occur.

2Pos219 タイプ 1 ロドプシンの L/Q スイッチがヘリオロドプシンの波長制御に及ぼす影響
Effects of the L/Q switch on color tuning of heliorhodopsin

Yuta Nakajima, Hideki Kandori (*Grad. Sch. Eng., NIT*)

Amino acid residues around retinal determine color of rhodopsins, which has been extensively studied experimentally and theoretically. Characteristic color-determinant residues are often called “switch”. L/Q switch is famous in type-1 rhodopsins, where green- and blue-absorbing proteorhodopsins (PRs) possess Leu and Gln at position 105, respectively. Previous mutation study revealed that volume of residues, not hydrophobicity, is the determinant of color in PR (Ozaki et al. 2014). Heliorhodopsins (HeRs) are the new rhodopsins, whose sequences highly differ from type-1 rhodopsin (Pushkarev et al. 2018). Comprehensive mutation study of the corresponding position in HeR and the comparison with type-1 rhodopsin will be presented and discussed.

2Pos220 クリプト藻由来のカチオンチャネルロドプシン Gt_CCR4 のオプトジェネティクスに向けた電気生理学的研究
Study of cation channelrhodopsin Gt_CCR4 from cryptophyte for optogenetics

Shunta Shigemura[1], Shoko Hososhima[1], Hideki Kandori[1], Satoshi Tsunoda[1,2] ([1]*Grad. Sch. Eng., NIT*, [2]*JST PRESTO*)

Gt_CCR4 is a novel light-gated cation channelrhodopsin from a cryptophyte algae Guillardia theta. Because a proton donor D96 of a H+ pump bacteriorhodopsin (BR) is conserved, Gt_CCR4 has close sequence homology to BR rather than the well-known Cr_ChR2. Thus, the channel gating and ion transport mechanism of Gt_CCR4 could be different from Cr_ChR2. In fact, Gt_CCR4 exhibits large channel current without significant inactivation and desensitization. We also demonstrate high Na+ selectivity of Gt_CCR4 in which selectivity ratio for Na+ is 37 fold larger than that for Cr_ChR2, promising an effective neuronal excitation without unfavorable pH change. Further mutation studies to improve the channel properties will be presented.

2Pos221 中赤外レーザーの神経系培養細胞のカルシウム濃度および膜電位への影響
The influence of mid-infrared laser on Ca2+ concentration and membrane potential of neuron-like cells

Yoshiyuki Shimizu, Toyohiko Yamauchi, Tatsuo Dougakiuchi, Gen Takebe (*Hamamatsu Photonics K.K.*)

Previously, we reported that irradiation of cultured cells with mid-infrared lasers can control intracellular Ca2+ concentration. We are paying attention to changes in the membrane potential of nerve cells among many physiological functions involved with calcium. In this study, cells derived from differentiated neurons (NG108-15) were irradiated with lasers to observe the reaction. We could increase intracellular Ca2+ concentration temporarily or continuously by changing the laser power. As for membrane potential, the responses varied from cell to cell, and further verification is required to identify the irradiation conditions that control them stably.

2Pos222 中赤外光照射による細胞のアポトーシスシグナル誘導
Induction of intracellular apoptotic cell signaling by mid-infrared laser exposure

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In this study, we investigated whether mid-infrared light with a wavelength of 6.1 micrometers can initiate apoptotic signaling in HeLa cells by observing caspase activity and mitochondrial membrane potential, both of which are known as markers for apoptotic cell analysis. The mid-infrared light was incident on the cell samples from the bottom of the dish, and fluorescence imaging was simultaneously performed using an upright fluorescence microscope. We confirmed from both markers that the progression of apoptotic cell death was triggered immediately after the laser exposure with a single irradiation of 0.1 s. This result indicates a high possibility that mid-infrared light can regulate cell viability.

2Pos223 生体エネルギーを浪費する光駆動内向きプロトンポンプロドプシンの電気生理学研究と光遺伝学への応用
Electrophysiological study and optogenetics application of inward-directed proton-pumping rhodopsin, NsXeR

Satoshi Tsunoda[1,2], Shoko Hososhima[1], Hideki Kandori[1] ([1]*Nagoya Institue of Technology*, [2]*JST PRESTO*)

All the light-driven ion pumps such as bacteriorhodopsin have been believed to maintain electrochemical potential which provides energy for most biochemical reactions. Thus, all known proton pumping rhodopsins have been outwardly directed. However we and others recently identified light-driven inward-directed proton pumping rhodopsins from marine bacteria, PoXeR and NsXeR. These pumps waste electrochemical potentials by actively transporting protons. Here we performed patch-clamp recording to characterize the pumping function of NsXeR. We demonstrate that NsXeR can transport protons against backward potential up to 160 mV. Extracellular pH affects the pumping rate. Mutation study around proton transport pathway and optogenetics experiment will be presented.

2Pos224 Photo-regulate small GTPase Ras using photochromic peptide inhibitor

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Small GTPase Ras is a central regulator of cellular signal transduction processes. The function of Ras is regulated by GEFs and GAPs. Previously, we have shown that GDP-GTP exchange of Ras with SOS is photo-regulated by peptide inhibitor crossliked with azobenzene-di-maleimide (ABDM). In this study, we employed di-iodoacetamide azobenzene (DIAAB) derivative to control GDP-GTP exchange. DIAAB incorporated into the SOSαH peptide showed cis-trans isomerization accompanied by UV and VIS light irradiations. CD spectrum analysis indicated that trans state of DIAAB-peptide showed αhelix secondary structure partially. On the other hand, cis state of the peptide showed random secondary structure. Effect of the photochromic peptide for GDP-GTP exchange of Ras was examined.

2Pos225 プリオンペプチド銅錯体のレドックスポテンシャル
Redox potential of copper-binding prion peptide

Shuhei Murakami, Wakako Hiraoka (*Grad.Sch.of Sci.& Tech.,Meiji Univ*)

Metal chelation to prion protein (PrP) is important to keep the physiological structure of PrP. Because recent reports show that the metal-reservoir function of PrP may be involved in redox homeostasis, we focused on the redox potential of octapeptide (PHGGGWGQ) acting as the copper-binding site located in the octarepeat region of human PrP. ESR combined with spin trapping showed that octapeptide modulated ROS obtained from copper divalent cation. When cultured cells were incubated with copper and octapeptide or antennapedia (cell penetrating peptide) connecting octapeptide, copper induced cytotoxicity was observed only in octapeptide without antennapedia treated cells. These results suggest the antioxidant potential of intracellular octapeptide against harmful metals.

2Pos226 低酸素下での X 線誘発 DSB の修復効率
Rejoining efficiency of X-ray-induced DSBs in hypoxia

Ryoichi Hirayama, Akiko Uzawa, Yoshiya Furusawa, Sumitaka Hasegawa (*NIRS, QST*)

Hypoxia suppresses the radiosensitivity to be one-third for X-rays than in normoxia. Radiosensitizing effect by oxygen is important, because it presents before, during and after the irradiation in the cell. However, the effects of oxygen on DNA DSB after irradiation are not well known. We investigated amount of DNA-DSB after rejoining under aerobic or hypoxic conditions, which produced in X-irradiated mammalian cells in hypoxia. The yield of DNA DSB was quantified by a static-field gel electrophoresis for each repair time. Rejoining of the DNA-DSB under hypoxic condition was slower (~1.5 times) and left more DNA fragments (~4 times) as compared to under aerobic condition. In summary, DSB rejoining is suppressed by the oxygen environment.

2Pos227 低線量放射線に曝露された細胞の運命決定における ATM を介した細胞質の放射線応答重要性
Importance of ATM-mediated cytoplasmic radiation response in determining the fate of cells exposed to low-dose radiation

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Studies with an X-ray microbeam cell irradiation system revealed that cell death is enhanced in nucleus-irradiated cells rather than whole irradiated cells in low-dose region. Considering the localization of deposited energy in irradiated cells, irradiation to the cytoplasm might induce certain type of intracellular signaling and thereby enhance cellular repair capability. From the gene expression analysis, it was suggested that in absence of irradiation to the cytoplasm, no transfer of ATM from cytoplasm to cell nucleus (nucleo-shuttling) is induced, so that sufficient DNA damage repair ability is not induced at low doses. ATM-mediated cytoplasmic radiation response is considered to play an important role in determining the fate of cells exposed to low-dose radiation.

2Pos228 酸化ストレスが引き起こす HeLa 細胞ミトコンドリア電子伝達系の機能増幅
Oxidative stress-induced enhancement of mitochondrial electron transport chain in HeLa cells

Wakako Hiraoka, Shuhei Murakami (*Department of Physics, Meiji University*)

A mitochondrial electron transport chain (ETC) plays an main role in energy metabolism in wide variety of biological systems. Analysis of enzyme activities and kinetic profiles of ETC is essential for assessing mitochondrial function to maintain homeostasis and prevent metabolic dysfunction. Our goal is to detect ETC enzyme activities in whole tissue or dissociated cells with electron spin resonance (ESR). ESR spectra of HeLa cells treated with hydrogen peroxide showed the increase of signal intensity of Fe-S, complex I, and semi quinone radical. Our results suggest that the enhancement of ETC enzyme activities should contribute to maintain a mitochondrial function against the redox disturbance such as oxidative stress.

2Pos229* DNA を自発的に取り込んだ細胞サイズ液滴
Aqueous polymer solutions create stable cell-sized sphere entrapping DNA: A novel scenario of de novo cell

Fumika Fujita[1], Hiroki Sakuta[1], Kanta Tsumoto[2], Takahiro Kenmotsu[1], Kenichi Yoshikawa[1] ([1]*Facul. Life Med. Sci., Doshisha Univ.*, [2]*Facul. Eng., Mie Univ.*)

Living cells maintain their lives through the construction of micro-compartment entrapping crowding macromolecules. Recently, we have found that micro-droplets generated in aqueous solution containing different soluble polymers uptake bio-macromolecules, such as DNA and actin and that these micro-droplets are rather stable by avoiding the fusion. In the present study, we have evaluated the stability of the cell-sized droplets entrapping λ-DNA (49kbp) generated through simple mixing of aqueous solution containing hydrophilic polymers, such as polyethylene glycol (PEG) and dextran (DEX). It was discovered that DEX-rich droplets entrapping-DNA remain with the sizes of several tens μm, suggesting the generation of self-organized de novo cell.

2Pos230* 遺伝子破壊変異の頻度が調節可能な系の構築およびそのゲノム縮小の進化実験への応用
Construction of a Genetic Tool for Tuning Gene-Inactivating Mutations and its Application to Experimental Evolution of Genome Reduction

Yuki Kanai[1], Saburo Tsuru[2], Chikara Furusawa[2,3] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*UBI, Univ. Tokyo*, [3]*BDR, RIKEN*)

To understand the evolution of life, causal study to prove tendencies in genomic evolution predicted by comparative genomics is important. However, it is generally difficult because the evolutionary rate in nature is sluggish. Here, we designed a mutator strain of Escherichia coli, which achieves high mutation rate utilizing the Tn5 transposon system. The strain appears to maintain high viability regardless of intensified mutation rate. As this property is suitable for experimental evolution, we believe this would be a first step towards future experimental studies of genomic evolution such as predicted genome reduction in eukaryogenesis. We expect we could present the result of an ongoing experimental evolution at the meeting.

2Pos231 Membraneless Polyester Microdroplets as Primordial Compartments at the Origins of Life

Tony Z Jia[1], Kuhan Chandru[1], Yayoi Hongo[1], Rehana Afrin[1], Tomohiro Usui[1], Kunihiro Myojo[2], Po-Hsiang Wang[1], H. James Cleaves[1] ([1]*Earth-Life Science Institute, Tokyo Institute of Technology*, [2]*Tokyo Institute of Technology Department of Earth and Planetary Science*)

Prebiotic chemistry was significantly diverse, and primitive living systems may have started from non-lipid boundary systems. Here, we show membraneless compartmentalization from prebiotically available α-hydroxy acids (αHAs). Membraneless microdroplets generated from polyesters synthesized from drying αHAs can segregate dyes and RNA and preserve biopolymer function, providing readily-available compartments that could facilitate chemical evolution by protecting, exchanging, and encapsulating primitive components. Facile polymerization of αHAs provides a novel pathway for the assembly of combinatorially diverse primitive compartments on early Earth, plausibly more accessible in a messy prebiotic environment.

2Pos232 情報高分子と連携したベシクルの自己生産
Reproduction of Vesicles coupled with Template Polymerization

Minoru Kurisu[1], Harutaka Aoki[1], Takehiro Jimbo[1], Yuka Sakuma[1], Masayuki Imai[1], Sandra Luginbuhl[2], Peter Walde[2] ([1]*Grad. Sch. Sci., Univ. Tohoku*, [2]*dep. Material, ETH*)

Molecular assembly system that have autonomous reproduction ability can be considered as minimal cell-like systems, which bridges non-living and living forms of matter. Here we show the reproduction of cell-sized vesicles coupled with polymerization on the surface of vesicles. The particular reaction used is the template polymerization of aniline occurring on the surface of AOT vesicles, which yields polyaniline emeraldine salt form (PANI). When AOT micelles are microinjected to AOT vesicles during polymerization, the AOT - PANI-ES vesicles selectively incorporate them in their membrane, which leads to a growth of the vesicle. If the AOT vesicles contained cholesterol, the vesicle not only showed growth, but also division, i.e., reproduction of vesicles.

2Pos233 Laboratory evolution of Escherichia coli reveals constrained evolutionary states for antibiotic resistance

Junichiro Iwasawa[1], Tomoya Maeda[2], Takaaki Horinouchi[2], Chikara Furusawa[1,2,3] ([1]*Dept. of Physics, Univ. of Tokyo*, [2]*RIKEN BDR*, [3]*UBI, Univ. of Tokyo*)

Understanding constraints which shape antibiotic resistance is key for predicting and controlling drug resistance. Here, we performed high-throughput laboratory evolution of Escherichia coli. By analyzing the genotype, gene expression and drug resistance data through interpretable machine learning techniques, the emergence of low dimensional phenotypic states was observed. Furthermore, by introducing commonly identified mutations to the parent strain, we successfully revealed the underlying biological processes responsible for the distinct states. These findings bridge the genotypic, gene expression, and drug resistance space, and lead to a comprehensive understanding of constraints for antibiotic resistance.

2Pos234 Fitness landscape of antibiotic-resistance evolution

Masayoshi Hiranaka[1], Nen Saito[2], Chikara Furusawa[2,3] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*UBI, Univ. Tokyo*, [3]*BDR, RIKEN*)

Bacteria can acquire multi-drug-resistance when exposed to various antibiotics. Recent studies using laboratory evolution experiments have revealed constraints in evolutionary dynamics of drug resistance. However, the emergence of such constraints is yet to be understood in the framework of fitness landscape. The difficulty underlying in this problem is that the fitness landscape can change depending on the type and the strength of the environmental stress. In this study, we present a simple abstract model which enables us to analyze evolutionary dynamics in a changeable fitness landscape. Using this model, we reveal the constraints imposed on the evolutionary transition between different resistant mutants, and discuss the controllability of resistance evolution.

2Pos235 Relationship between fluctuation of single-enzyme activity and evolvability

Hiroshi Ueno[1], Morito Sakuma[1], Yoshihiro Minagawa[1], Kazuhito Tabata[1], Kentaro Miyazaki[2,3], Hiroyuki Noji[1] ([1]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*, [2]*AIST*, [3]*Grad. Sch. Front. Sci., Univ. Tokyo*)

Recently, the evolutionary fluctuation-response relationship has been proposed, where genes with larger phenotypic fluctuation have higher evolution speed (Kaneko and Furusawa, 2006). However, it is not obvious whether this theory can be applied to any phenotype. Our recent studies have shown that there is a fluctuation in the activity of single enzymes synthesized from the same DNA. So, to elucidate the relationship between “fluctuation of single-enzyme activity” and “evolvability”, directed enzyme evolution and the measurement of single-enzyme activity were conducted using femtoliter reactor array device. We succeeded to obtain several high active mutants and, interestingly, found that fluctuations of single-enzyme activity were different depending on the mutant.

2Pos236 数理モデルとライブイメージングデータを用いた分裂酵母間期核内構造の解析
Analysis of fission yeast interphase intranuclear structure by mathematical model and live imaging data

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Eukaryote genome dynamics are closely related to intra-nuclear functions such as transcriptional regulation and DNA repairs. Fission yeast genome was studied as a model of eukaryote interphase chromosome. Recent mathematical models of them assumed that spindle pole body (SPB) anchoring all centromeres was always located at the opposite of nucleolus. However, recent live imaging studies suggested SPB moves dynamically along the microtubule. Additionally, they showed the nucleolus and nuclear membrane change their shape and positional relationships. In this study, we constructed a realistic model of interphase intranuclear structures of fission yeast based on the estimation of relative positions and orientations among SPB, nucleolus, and nucleus from live imaging data.

2Pos237 Dynamical chromatin organization during transcription

Ashwin S. Selvarajan[1], Kayo Hibino[2], Yuji Itoh[2], Kazuhiro Maeshima[1,2], Masaki Sasai[2] ([1]*Dept of Applied Physics, Nagoya University, Nagoya, Japan*, [2]*Structural Biology Center, National Institute of Genetics, Mishima, Shizuoka, Japan*)

Understanding the link between genome organization and gene regulation is a challenge in molecular biology. It was recently found that chromatin movement in the interphase nucleus is globally enhanced when transcription is suppressed, suggesting that interactions of transcriptionally active RNA polymerase II (RNAPII) with droplets/clusters of transcription factors and cofactors impose a global constraint on the chromatin movement; addition of RNAPII inhibitor such as DRB downregulates this constraint and upregulates the chromatin dynamics. Using single nucleosome tracking data from live human cells and simulation/theoretical techniques, we study the effects of the constraint on the displacement distribution of nucleosomes and contact maps between chromatin regions.

2Pos238 機械学習を用いたタンパク質部分配列の構造及び機能の解析
Analysis of structural and functional propensities for subsequences of proteins by using machine learning

Ryohei Kondo[1], Kota Kasahara[2], Takuya Takahashi[2] ([1]*Grad. Sch. Life Sci., Ritsumeikan Univ.*, [2]*Coll. Life Sci., Ritsumeikan Univ.*)

Toward elucidation of principle of protein sequence-structure-function relationship, analyzing the enormous data of protein structure accumulated in Protein Data Bank (PDB) is crucial. In this study, we focused on the nature of subsequences rather than whole sequences to understand the relationship of sequences and structures. Specifically, we decomposed sequences of many proteins in the PDB into subsequences consisting of N successive residues termed N-grams, where N denotes the number of residues. We predicted whether a subsequence is in an intermolecular interface or not, secondary structure, and accessible surface area (ASA) of N-grams by using Artificial Neural Network. In result, we predicted a possibility of intermolecular interaction of 6-grams as about 69 %.

2Pos239 GPCR ー G タンパク質の結合選択性に関わる部位同定および結合 G タンパク種予測への応用
Determination of key regions relating GPCR-Gprotein coupling selectivity and their application for predicting coupling G-protein kinds

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For the drug discovery, it is important to predict G protein kinds activated by a GPCR-ligand combination. We intend to identify key sites relating to G protein kinds, using structure/sequence information. Ligand-GPCR-G protein combination were obtained from public DBs. We made 9 comparative model of adrenergic receptor families, and computed the interaction profile of them by MD simulation. The physicochemical parameters of ligand, ligand binding sites and interaction profile were unified as vectors. We evaluated the relationship of 15 kind of G-protein selection and 934 vectors using SVM method. The high prediction accuracy of intersection official approval (94.4%) and the objective test (95.0%) suggested the essential contribution of the interaction profiles.

2Pos240 デノボデザインによる新規 αβ 型蛋白質フォールドの探査
Exploration of novel alpha-beta protein folds by de novo design

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Naturally occurring protein folds are only an infinitesimal subset of the considerable protein fold patterns. Why are there nonexistent folds in nature? These novel folds might be physically un-foldable or left undiscovered in evolution. Here, we attempted to address the reason by designing the novel fold proteins. PDB analyses with a set of rules of β-sheet topology identified a comprehensive set of physically feasible novel β-sheet topologies. Then, structures for all of the novel 4-stranded topologies (8 folds) were computationally designed. The designs, expressed in E.coli and purified, are found to be stable in solution for all the 8 folds. The NMR structures of designs for the 4 folds have been determined so far, showing close agreement to the design models.

2Pos241 アンサンブルドッキングによるタンパク質の相互作用面の解析
Analysis of protein interaction surfaces using ensemble rigid-body docking process

Nobuyuki Uchikoga[1], Yuri Matsuzaki[2] ([1]*Dept. of Network Design, Sch. of Interdiscip. Math. Sci., Meiji Univ.*, [2]*ToTAL, TITech*)

For predicting protein-protein interactions using rigid-body docking methods, it is not necessary to know information of protein interaction surfaces explicitly because there are many softwares for predicting interaction protein pairs with high accuracy. On the other hand, to understand a mechanism of protein-protein interactions, we need to know properties of protein interaction surfaces. Then, we have developed a method of interaction fingerprints (IFPs) for analysis of protein-protein interaction mechanisms. In this work, we examined ensemble rigid-body docking method for investigating protein interaction surfaces. Ensemble protein structures were generated from molecular dynamics process and docking surfaces of structures were analyzed using IFPs.

2Pos242 転写因子 Med26 における天然変性蛋白質認識メカニズムの分子動力学的検討
Molecular dynamics study for elucidation of recognition mechanism of intrinsically disordered proteins by transcription factor Med26

Satoshi Goto[1], Takuya Takahashi[2], Kouta Kasahara[3] ([1]*Coll Life Sci., Ritsumeikan Univ*, [2]*Prof. Univ.Ritsumei*, [3]*Ass.Prof. Univ.Ritsumei*)

Elucidation of the molecular mechanisms of transcription is a central issue in biophysics. In particular, how mediator complex recruits different transcription factors depending on the target gene is not well understood. Recently, it has been found that the transcription factor Med26 functions as a molecular switch by interacting intrinsically disordered proteins TAF7 and EAF1. However, the microscopic mechanism of how Med26 recognizes TAF7 and EAF1 remains unknown at the atomic resolution. Therefore, in this study, we investigate the mechanism of recognition of TAF7 and EAF1 by Med26 by multi-canonical MD simulations. Resultant free-energy landscapes showed high conformational heterogeneity of ligands. That suggest fuzzy complex formation for their recognitions.

2Pos243* (2SCP-6) 上皮メカノケミカル動態の同定
(2SCP-6) System identification of mechano-chemical epithelial sheet dynamics

Yoshifumi Asakura[1], Yohei Kondo[2], Kazuhiro Aoki[2], Naoki Honda[1] ([1]*Grad. Sch. Biostudies, Univ. Kyoto*, [2]*Div. Quantitative Biol. ExCELLS, NIBB.*)

Collective migration of epithelial cells is a fundamental process of multi-cellular organisms. Our recent study using live imaging with FRET-based biosensor discovered that cell migration within an epithelial sheet is oriented by traveling waves of ERK activation. However, how the cells make a decision on migration direction by integrating mechano-chemical signals has remained still elusive. Here, we performed reverse-engineering approach to extract a hidden control role in the epithelial sheet dynamics in a data-driven manner. Our model has an ability to forecast cell migration quantified in time-lapse images. Therefore, our approach would be powerful to understand mechano-chemical epithelial sheet dynamics.

2Pos244* 細胞周期の不均一性に関する網羅的数理モデルの構築
A comprehensive model of heterogeneous cell cycle responses

Hiroaki Imoto, Kyouichi Ebata, Shigeyuki Magi, Suxiang Zhang, Mariko Okada (*IPR, Osaka Univ.*)

The principles governing cellular decision-making are poorly understood. Because many cells show similar quantitative effect of signaling activities on downstream gene expression, we hypothesized distinct dynamics of transcription factors among cell types might stem from a universal network mechanism. To reveal general mechanisms underlying cell system regulation, we constructed a comprehensive mathematical model describing the processes of signal transduction, transcriptional regulation and cell-cycle progression. We found that the heterogenous responses can be explained by different parameter sets in the common network structures. By comparing parameter sensitivities, we revealed ERK activation dynamics plays a critical role in generating cell-type specific responses.

2Pos245* 真性粘菌変形体の走磁性とその探索行動への寄与
Magnetotaxis of Physarum plasmodium and its contribution to exploration

Michinori Muro, Hiroshi Sato, Tomohiro Shirakawa (*Natl Def Acad Japan*)

The plasmodium of Physarum polycephalum is a unicellular and multinuclear giant amoeba, and has many kind of taxes such as chemotaxis, phototaxis, thermotaxis, and so on. Since the plasmodium also has electrotaxis, we predicted that the organism has a reactivity on electromagnetism, and thus that it has magnetotaxis. In our experiment, we actually found that the plasmodium has magnetotaxis. The plasmodium moves along the magnetic lines, and chooses its direction of the move in north or south depending on the strength of the magnetic field and its body weight. In our simulation, we further proved that such properties of the magnetotaxis contributes to the exploration by the organism.

2Pos246 (2SHP-5) 細胞内のインスリン様成長因子-I（IGF-I）シグナルは振動する
(2SHP-5) Cellular insulin-like growth factor-I (IGF-I) signal can be oscillated

Masato Masuda, Fumihiko Hakuno, Shin-Ichiro Takahashi (*Dep. App. Ani. Sci., Grad. Sch. Agr. Lif. Sci., The Univ. Tokyo*)

Insulin-like growth factor (IGF)-I, whose structure is similar to proinsulin, is well known to be secreted constantly, and to mediate long-term actions such as cell growth, differentiation and survival. Recently we have reported studies on the detailed regulatory mechanism of IGF-I signaling including feedback loop. Based on these results, we made a numerical model to simulate the IGF-I signaling in the constant presence of IGF-I. Simulation results and Routh-Hurwitz stability criterion of this model indicated that the IGF-I signal is shown to be stable when IGF-I concentration is high, whereas the IGF-I signal is oscillated when IGF-I concentration is low. Our analysis demonstrated that IGF-I signal can be oscillated in each cell in the constant presence of IGF-I.

2Pos247 BOID における第 4 のルール：他個体への注目は一定時間毎に一定の確率で失われる
The fourth rule of BOID: attention to the other individuals is lost with fixed probability every fixed time

Tomohiro Shirakawa, Hiroshi Sato, Takuya Matsuo (*Natl. Def. Acad. Japan*)

Raynolds's BOID model is an established model of swarm that well duplicates swarm behavior. However, in BOID swarm, the relative position of the individuals are almost fixed, though in real swarm the individuals actively exchange the positions each other. BOID fails to reproduce the fluctuation inside the swarm. To understand the mechanism of the internal fluctuation of swarm that BOID fails to reproduce, we performed observation on the swarm of tropical fish. In our experiment, we analyzed the duration time on how long each pair of two individual stay the neighborhood. As a result, our data implied that, in the fish swarm, attention to the other individuals is lost with fixed probability every fixed time.

2Pos248 An Information-theoretic approach toward identifying the leader(s) and aggregation place in Dictyostelium Discoideum colony

Udoy Sankar Basak[1], Sulimon Sattari[1], Kazuki Horikawa[2], Tamiki Komatsuzaki[1] ([1]*Hokkaido University*, [2]*Tokushima University*)

Dictyostelium Discoideum behaves as single and independent cells under some conditions but when they are challenged by adverse conditions, e.g., starvation etc., they get aggregated and form a multicellular structure. Our aim is to quantify the cell-to-cell relationship from the cell motility and cAMP signals and explore a singularity cell that may conduct the surroundings. We also want to predict the aggregation place in advance. Transfer entropy (TE) can be used to measure information flow between two processes. In case of leader and follower relationship, information transferred from a leader cell to follower cells should be greater than from a follower cell to a leader cell. To identify the aggregation place we apply particle imaging velocimetry (PIV).

2Pos249 細胞内共生進化への理論的アプローチ
Theoretical approach to evolution of intracellular symbiosis

Sakura Aoki, Kunihiko Kaneko (*The University of Tokyo Graduate School of Arts and Scienses*)

Intracellular symbiosis is an important factor to understand the evolution of diversity and complexity of life. However, the mechanism of evolution is poorly understood. It is not obvious that how a host and its internal organisms can maintain intracellular symbiotic relationship through evolution. To answer this, we considered a simple model of intracellular symbiosis among host cells and symbionts, which is internal subcellular organisms in hosts and not distinguished from parasites here. In this model, host cells and symbionts exchange materials necessary for growth. We analyzed the behavior of the dynamics for establishing this symbiotic relationship and uncovered its distinctive features.

2Pos250 Asymptotic expansion of a stochastic FitzHugh-Nagumo model

Takanobu Yamanobe (*Sch. Med., Hokkaido Univ.*)

We calculate an asymptotic expansion of the transition density of the FitzHugh-Nagumo model including a diffusion process. The first order term of the expanded solution process is Gaussian. However, since the deterministic version of the FitzHugh-Nagumo model and the backward fundamental solution of the Jacobi equation cannot be solved analytically, this leads to difficulties to calculate the variance-covariance matrix. We overcome these difficulties by combining the numerical solution of the deterministic FitzHugh-Nagumo model, the numerical backward fundamental solution of the Jacobi equation and Romberg integration scheme. We also compare the asymptotic expansion of the transition density with the one by Monte Carlo simulation to check the validity of the expansion.

2Pos251 光応答性を持った C. elegans の集団運動
Collective motion of optically susceptible C. elegans

Ken Nagai[1], Hiroshi Ito[2], Takuma Sugi[3] ([1]*JAIST*, [2]*Kyushu Univ.*, [3]*Shiga Univ. Med. Sci.*)

When number density is large enough, C. elegans collectively forms dynamical network comprising many bundles of worms on a substrate. Using a multi-particle model, we have found that the key factors to form the collective motion are long-time memory of rotation rate and short-range alignment interaction of worms. To investigate response properties of the collective motion to external stimuli, we endowed worms with optical susceptibility using optogenetics. One of the major responses caused by the mutation is activation of halted worms by optical stimulation. By uniform illumination of blue light, bundles of the mutant worms collapsed. Simulations of our multi-particle model suggest that the increase of active worms caused the bundle break.

2Pos252 逆ミセルを用いたフレーリッヒ凝縮の研究 (I)
Studies of Frohlich condensation using reverse micelles. I

Hiroshi Murakami (*QST*)

Frohlich proposed a theoretical model of cells regarding cells as dielectrics. The model shows that a condensation takes place at the lowest frequency mode of polar vibrations in a terahertz range if a metabolic energy exceeds a threshold. Experimental studies using cells supported the validity of the model, but did not verify it directly because of complexity of cells. We here use reverse micelles as a simplified model of cells. Reverse micelles are microscopic water droplets covered by a membrane formed by a self-assembly of surfactant molecules, and a hydrophilic molecule can be dissolved in the droplet. Further, we use a light energy in place of a metabolic one, that is, a dye molecule is introduced in a reverse micelle to convert a light energy to a thermal one.

2Pos253 孤立した粒子の運動解析を基とした、回転する自走粒子の集団運動を表す数理モデルの推定
Estimation of mathematical model representing collective motion of rotating self-propelled particle

Tadashi Sakaguchi[1], Kazuhiro Oiwa[2], Hitoshi Sakakibara[2], Ken Nagai[1] ([1]*JAIST*, [2]*NICT*)

Collective motion, where numbers of self-propelled organisms form a large-scale structure by interacting each other, is widely seen in nature, such as bird flocks and fish schools. To demonstrate collective motion by particles with local aligning interaction in in vitro environment, motility assay is commonly used, in which microtubules, rod-like particles, are propelled by molecular motors adhered to a substrate. Using dynein c, each microtubule draws turning tracks, and with a high density of microtubules, they form vortex lattice (Sumino, et.al, 2012). In this research, we observed tracks of isolated self-propelled microtubules and measured the variation of curvature in long time scale to estimate the best model of collective motion in motility assay.

2Pos254 抗原識別における確率的ノイズの役割
A role of stochastic noise in ligand discrimination

Masashi K. Kajita, Kazuyuki Aihara, Tetsuya J. Kobayashi (*IIS, University of Tokyo*)

Intracellular reactions are inherently stochastic processes. Nonetheless, cells can accurately sense and respond to an environmental signal. Conventional theories of cellular decision making have revealed the decoding mechanisms. However, the ubiquity of structurally-similar non-target molecules in the environment can hamper appropriate information-transduction by non-specific interaction. In this research, we address the problem of how cells accurately distinguish the target molecules by proposing a mathematical mechanism, which can accurately discriminate the target and non-target molecules in a noisy environment. We also discuss a possible connection between the mechanism and the phenomena recently found by single-cell imaging.

2Pos255 Theoretical model of dynamics of epithelial tissue with cellular chirality

Takaki Yamamoto[1], Tetsuya Hiraiwa[2], Tatsuo Shibata[1] ([1]*RIKEN BDR*, [2]*Mechanobiology Institute, National University of Singapore*)

The morphology of organisms, organs and tissues often breaks chiral symmetry. Understanding the multicellular dynamics is essential to elucidate the mechanism. In contrast to such multicellular chirality, single-cell chiral behavior has been recently reported as the unidirectional rotation of isolated cells. From the view point of mechanics, the observations imply that the single cells can cell-autonomously generate torques. Here, we ask a question how the torque generated by single cells affects the multicellular dynamics, and if it can induce chiral morphogenesis. To tackle this problem, we propose a theoretical model of multicellular dynamics with torques generated by single cells. We report how the cellular torque affects the multicellular dynamics.

2Pos256* 細胞内温度場は高分子に依存する
Intracellular temperature field depends on polymers

Masaharu Takarada[1], Kohki Okabe[1,2], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ of Tokyo*, [2]*PRESTO, JST*)

Previous studies have suggested that intracellular temperature changes spatiotemporally about 1K. This temperature change inside cells is significantly larger than the temperature change simulated assuming that the thermal properties of the cytoplasm is water. Here, we hypothesized that biopolymers are responsible for this difference. To prove this, we tracked transient change in intracellular temperature by using an infrared laser-based heating system and a fluorescent polymer thermometer. Temperature dynamics in local area of the cytoplasm allowed for a quantitative evaluation of heat transfer in cells, revealing that while some polymers confine heat, others broaden localized heat. These results showed that biopolymers create unique temperature field in cells.

2Pos257* インフルエンザウイルス多様性解析に向けた、多次元デジタル計測技術の開発
Multi-Dimensional (MD) digital assay for analysis of influenza virus heterogeneity

Shingo Honda[1], Yoshihiro Minagawa[2], Kazuhito V. Tabata[2], Hiroyuki Noji[2] ([1]*Dept. Bioeng., Grad. Sch. Eng., Univ. Tokyo*, [2]*Dept. Appl. Chem., Grad. Sch. Eng., Univ. Tokyo*)

Neuraminidase (NA) is an enzyme essential for the infection of influenza virions and known to obtain resistance against its inhibitors by genetic heterogeneity. To understand the heterogeneity of NA and its resistance in single-virion resolution, we established a Multi-Dimensional (MD) digital assay. In this assay, we can conduct serial NA activity assays with several doses of an inhibitor, for individual virions encapsulated in micron-sized reactors. This enables estimations of NA resistance against the inhibitor for each virion. We have successfully validated this assay with a model enzyme alkaline phosphatase. We are applying this assay for influenza virions and have already succeeded in encapsulating the virions into the reactors.

2Pos258* Development of a method for quantitative profiling of microRNAs in single exosomes

Cinya Chung, Ryo Iizuka, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

Exosomes are nanosized membrane vesicles secreted by various types of cells. Recent studies have shown that cancer cells secrete more exosomes than normal cells and the exosomes contain specific microRNAs (miRNAs). Thus, exosomes in body fluids could provide potential information for cancer cells. To enable earlier and more accurate cancer diagnosis, we devised a method for profiling of miRNAs from single exosomes. Using synthetic miRNAs, we have demonstrated the practical application. Details will be discussed.

2Pos259* 光ファイバーを用いた蛍光相関分光法の開発とエクソソーム研究への応用
Development of optical fiber based fluorescence correlation spectroscopy and application to exosome study

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Exosome is an extracellular vesicle released by a variety of cells and contains proteins and nucleic acids. It plays a role of signaling to distant cells and tissues. Exosome released from cancer cells exists in body fluid and it includes cancer-specific biomolecules. Therefore, detection of cancer-specific exosome is useful for cancer diagnosis. In this study, we developed microscope mounted type FCS unit (Fiber-connected FCS; FC-FCS). FC-FCS is much smaller and simpler than conventional FCS device. FCS measurement can be realized by connecting to the camera port of the microscope without any adjustment. FC-FCS measurement was performed using an exosome-specific fluorescently labeled antibody to compare the expression ratio of specific membrane proteins in exosome.

2Pos260* 細胞内温度変動に関与する分子のスクリーニング法の開発
Development of a method to screen molecules related to intracellular temperature variation

Takashi Mitsubori[1], Kohki Okabe[1,2], Fumi Kano[3], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*PRESTO, JST*, [3]*IIR, Tokyo Inst. of Tech.*)

Recent studies on intracellular temperature imaging using fluorescent polymeric thermometer (FPT) have revealed that there is spatiotemporal temperature variation in the intracellular local area. The purpose of this study is to reveal the contribution of various biomolecules to intracellular thermodynamics. We produced resealed COS7 cells in which biomolecules were effluxed and developed a method for thermometry using FPT in these cells. We found that the temperature increase caused by infrared laser irradiation or mitochondrial thermogenesis was reduced in resealed cells compared with that in normal cells. This suggests that biomolecules affect intracellular temperature variation by contributing to intracellular thermodynamics.

2Pos261 Optical tweezers with red laser for new applications

Tomohiro Masuda, Wataru Nakashima, Kazuki Nakajima, Shin Yamaguchi, Takashi Sagawa,
Yuichi Inoue (*SIGMAKOKI CO., LTD.*)

Light source of optical tweezers, typically ~1000 mW and ~1064 nm, has changed into a weak red laser (4.5 mW, 635 nm) with a combination of Core Unit components from SIGMA-KOKI. With this system, 1 μm-polystyrene bead could be captured using not only a typical high NA objective, but also dry objectives with NA0.5-0.8. The red laser system has several advantages as 1) negligible heating of bulk water due to low adsorption than infrared laser, 2) easy construction and low risk due to visibility of the laser, and 3) reduced initial cost to ~1/10 with keeping possibility of future upgrade. Therefore, the system can be used in new applications as carbon nanotube-based local activation of biomolecules (Inoue et al., 2015) as well as educational training.

2Pos262 赤外超解像顕微鏡によるガチョウおよびペンギン羽毛の内部構造観察
Orientation-sensitive molecular imaging of keratin proteins of goose and penguin feathers by an IR super-resolution micro-spectroscopy

Hirona Takahashi, Koki Kimura, **Makoto Sakai** (*Faculty of Science, Okayama University of Science*)

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). We have already reported that orientation-sensitive molecular imaging of β-keratin at the rachis of goose feather and revealed the distribution and orientation by an IR super-resolution micro-spectroscopy based on non-linear optical process. In this study, we applied this method to β-keratins of penguin feather. In the presentation, the results of polarization dependent measurement will be discussed in detail. Results for penguin feather will be compared with these of goose feather.

2Pos263 ミクロゲルの粘弾性特性の測定
Viscoelastic measurement of a biopolymer microgel by microcapillary aspiration

Atsushi Sakai[1,2], Yoshihiro Murayama[1], Miho Yanagisawa[2] ([1]*Tokyo Univ. of Agri. & Technol.*, [2]*Komaba Inst. Sci., The Univ. Tokyo.*)

Biopolymers microgels are indispensable for cell culture and biomedical materials. Control of their mechanical properties is important because they are closely related to the mechanical sensing of cells. The mechanical properties of microgel solutions have been studied using rheometer, etc. However, those of single microgels are still elusive because they are difficult to measure. By using microcapillary aspiration, we succeeded in measuring the elasticity of gelatin microgels and found that smaller microgels have higher elasticity than bulk gels (Sakai et al., 2018 ACS Cent. Sci.). In this presentation, we will introduce a method to measure microgel viscoelasticity by periodically aspirating the microgel. This should be applicable to live cells.

2Pos264* 高速イオン伝導顕微鏡による表面電荷のマッピング
Mapping of surface charge by high speed ion conductance microscopy

Shusei Kaihatsu[1], Kazuki Shigyo[2], Toshio Ando[2], Shinji Watanabe[2] ([1]*Grad. Sch. Math. & Phys., Kanazawa Univ.*, [2]*WPI-NanoLSI, Kanazawa Univ.*)

The heterogeneities of interfacial charge of plasma membranes are thought to play an important role in many cellular processes such as cellular uptake, cell growth and division, and adherence. Scanning ion conductance microscopy (SICM) is powerful tool to map local surface charge of plasma membranes of live cells with nanometer scale resolution among existing experimental techniques. However, the temporal resolution of charge-sensing SICM is quite low. Here, we demonstrated the improvement in the temporal resolution of charge-sensing SICM by analyzing current-distance approaching curves measured in close proximity to the sample surface. We will report the result of the application of our proposed method in supported lipid bilayer formed on mica substrate.

2Pos265* マニピュレーター付き高速 AFM スキャナーの改良
Improvement of high-speed AFM scanner with manipulator

Jun Takano[1], Shun Aoki[2], Kazuki Shigyo[3], Shinji Watanabe[3], Toshio Ando[3,4], Noriyuki Kodera[3,4] ([1]*Grad. Sch. Math. & Phys., Kanazawa Univ.*, [2]*Sch. Math. & Phys., Kanazawa Univ.*, [3]*WPI-NanoLSI, Kanazawa Univ.*, [4]*CREST, JST*)

Direct observation of proteins at work under an external load by high-speed AFM (HS-AFM) would greatly gain the mechanistic insight of their functional modulation under the load. To realize this observation, we have been developing a HS-AFM scanner with manipulator. Last year, we reported that the sub-μm sized objects can be moved by the end of a manipulator glass during HS-AFM imaging. Here, we improved the scanning performance of the manipulator by reducing a friction between the manipulator glass and an AFM substrate surface. In addition, the manipulator end was sharpened from ~200 nm to ~30 nm. These improvements allowed us to freely move F-actin while resolving their half helical pitches. We now apply this scanner system to actin and actin binding proteins.

2Pos266* タンパク質オリゴマー分布イメージング：生細胞内で空間的に不均一なオリゴマーの分布可視化に向けて
Protein Oligomer Imaging: towards Visualization of Spatially Heterogeneous Oligomer Distribution in Living Cell

Ryosuke Fukushima[1], Johtaro Yamamoto[2,3], Masataka Kinjo[2] ([1]*Grad. Sch. of Life Sci., Hokkaido Univ.*, [2]*Fac. of Adv. Life Sci., Hokkaido Univ.*, [3]*Biomed. Res. Inst., AIST*)

Protein oligomerization is a key to understand controlling mechanism in protein function. Number and brightness analysis (N&B) enable to visualize images of oligomer distribution over time in living cell by fluorescence signal statistics. However, its precision is not enough to distinguish a small change between monomer and oligomer. In this study, we applied maximum likelihood estimation (MLE) and two-detector system in order to improve precision and accuracy. MLE gave 1.3 times precise estimator. The two-detector system reduced the photon count loss caused by detector dead time, and it increased the accuracy. The improved N&B was applied to characterize oligomeric state of glucocorticoid receptor, which belongs to nuclear receptor superfamily and dimerize in nucleus.

2Pos267* A ratiometric bioluminescent indicator for water hardness in living specimen

Md Nadim Hossain[1,2], Ryuichi Ishida[2], Mitsuru Hattori[1,2], Tomoki Matsuda[1,2], Takeharu Nagai[1,2] ([1]*Graduate School of Engineering, Osaka University*, [2]*ISIR, Osaka University*)

Water hardness is defined as a sum of divalent metallic ions concentration in water, whose main elements are Ca2+ and Mg2+. It strongly affects on human health such as cardiovascular diseases, eczema, kidney stone, hypomagnesemia, and hypocalciuria. However, how the water hardness is involved in pathogenesis is unknown because there is no relevant indicator for measuring water hardness in the living specimen. Here, we report a FRET-based genetically encoded ratiometric bioluminescent indicator called LOTUS-W. It is sensitive to Ca2+ (Kd =2.1 mM) and Mg2+(Kd =4 mM) and enables to real-time monitoring of water hardness as bioluminescence color change in water. In the future, we will try to compare the difference of water hardness in normal and pathological tissue.

2Pos268 (2SHP-4) 大気圧走査電子顕微鏡 ASEM による骨組織再構築の水中免疫電顕法と cryo-TEM 観察
(2SHP-4) Observation of unstained bone tissues and immuno-EM in liquid by ASEM and cryo-TEM

Chikara Sato[1], Shinya Sugimoto[2], Yuri Hatano[1], Mari Sato[1], Eiko Sakai[3] ([1]*Biomedical Res. Inst., AIST*, [2]*Dept. Bacteriol., The Jikei Univ. Sch. Med.*, [3]*Dental Pharmacology, Nagasaki Univ.*)

ASEM observed aldehyde-fixed bone tissue mineralization in natural aqueous buffer, decreasing the risk of changes. Mineralization trafficking in osteoblasts and trabeculae of mineralized femur spongy bone surrounded by cortical bone were directly visible. Associated large and small cells revealed by PTA staining suggest remodeling by bone-forming osteoblasts and bone-absorbing osteoclasts, confirmed by immuno-EM in solution targeting cathepsin-K protease digesting collagen fibers in bone[1]. Axonal segmentation controlling neuron trafficking was visualized[2]. MRSA biofilm formation was monitored by ASEM[3] and cryo-TEM. [1]Sato et al. Scientific Reports 9, 7352, 1-13 (2019). [2]Kinoshita et al. Sci Rep 7,41455,1-14 (2017). [3]Sugimoto et al. Sci Rep 6,25889,1-13 (2016)

2Pos269 (2SEA-5) G1 期酵母細胞核内における核酸分布の XFELX 線回折イメージング
(2SEA-5) Distribution of nucleic acids in yeast nucleus of G1 phase visualized by X-ray diffraction imaging using X-ray free electron laser

Masayoshi Nakasako[1,2], Takahiro Yamamoto[1,2], Amane Kobayashi[1,2], Mao Oide[1,2], Koji Okajima[1,2], Yuki Takayama[1,2,3], Tomotaka Oroguchi[1,2], Masaki Yamamoto[2] ([1]*Keio University*, [2]*RIKEN*, [3]*University of Hyogo Prefecture*)

Distribution of nucleic acids in yeast nuclei was visualized by X-ray diffraction imaging using X-ray free electron laser. We obtained high quality diffraction patterns of frozen-hydrated yeast nuclei at the G1 phase. From the diffraction patterns, we first found mass and surface fractal properties with respect to the packing of substructural elements. The projection electron density maps retrieved form diffraction patterns indicated non-uniform distribution of nucleic acids including related proteins. From the maps, we speculated a model with a U shape to explain most of projection maps observed.

2Pos270 ホウレンソウグラナ膜における光化学系 II 分子間相互作用の高速 AFM による解析
HS-AFM imaging and analyses of intermolecular interaction of photosystem II in grana membrane from spinach

Daisuke Yamamoto (*Fuc. Sci. Fukuoka Univ.*)

Photosystem II (PSII) is a large protein complex involved in the photosynthetic reaction. The photon energy harvested by light harvesting complexes (LHCs) is funneled into PSII. While the structures of PSII-LHCII complexes have recently be solved at near-atomic resolution, the higher order structure of PSII-LHCII and its dynamic behavior in grana is elusive. Here, grana membrane from spinach was examined by HS-AFM, and the molecular architecture of PSII in its native habitat was analyzed. The dimer of PSII core was clearly observed, which enabled fitting of HS-AFM images of PSII to the high-resolution cryoEM structures. Analysis of the lateral diffusion of PSII suggested that some PSII-LHCII complexes associate each other and form large complexes in the membrane.

2Pos271 光照射でタンパク質機能阻害・細胞死を誘導する単量体光増感緑色蛍光タンパク質の開発
Monomeric green fluorescent protein based photosensitizer for photo-inducible protein inactivation and cell death

Tomoki Matsuda[1], Yemima Dani Riani[1], Kiwamu Takemoto[2], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*Grad Sch. of Med., Yokohama City Univ.*)

We developed a green fluorescent protein based photosensitizer SuperNova Green (SNG) that enables spatiotemporal control of ROS production upon blue light irradiation. The SNG produces inconsiderable amount of singlet oxygen and likely to produce superoxide and its derivatives. Light-dependent inactivation of the PH domain of phospholipase C-δ1 and induction of the death of HeLa cells were succeeded for the fusion protein with SNG and cells expressed mitochondria localized SNG, respectively. As a proof of concept for multiple control of different proteins or cell types, we demonstrate that SNG can be used with its red variant, SuperNova Red, to perform selective protein inactivation or cell death induction in a spatiotemporal manner by lights with different wavelength.

2Pos272 RNA ポリメラーゼ II により制御されるクロマチンダイナミクス
Chromatin dynamics regulated by RNA polymerase II

Yuji Itoh, Michael Babokhov, Kayo Hibino, Kazuhiro Maeshima (*NIG*)

Recent studies revealed dynamic behavior of chromatin in live cells. The chromatin dynamics is suggested to play an important role in the molecular processes on DNA like mRNA transcription. To investigate the interplay between transcription and chromatin dynamics, we fluorescently labeled histone H2B with a HaloTag ligand and tracked single nucleosome motion by an oblique illumination microscopy. The treatment of cells by a Cdk7 inhibitor enhanced the chromatin dynamics genome-wide, whereas the treatment by a Cdk9 inhibitor did not. We found that the phosphorylation of Ser5 in the C-terminal domain of RNA polymerase II is well correlated with the chromatin dynamics and thus propose a model in which the transcription initiation by RNAPII regulates the chromatin dynamics.

2Pos273 電子顕微鏡トモグラフィ像への構造フィッティング
Fitting atomic structure to Electron Microscopy Tomography

Yuki Mori, Suguru Kato, Toru Niina, Shoji Takada (*Kyoto University*)

Electron microscopy tomography (EMT) can observe macromolecules in cellular environment with molecular-resolution. For example, ChromEMT, EMT with a labeling method, succeeded to observe chromatin structure at nucleosome resolution in human cells. While there are several approaches to model atomic structure into the observed Cryo-EM maps, they are mainly aiming to fit one or a few structures into the single particle analysis data. However, EMT image contains a large number of molecules so that previously-developed methods are not suitable. In this study, we examine the new method to fit structures roughly to maps with rigid body fitting followed by the flexible fitting.

2Pos274 高輝度なポジティブ型光スイッチング蛍光タンパク質 Kohinoor 2.0 の開発
Development of a highly-bright positively reversibly photoswitchable fluorescent protein Kohinoor 2.0 for super-resolution microscopy

Tetsuichi Wazawa[1], Shusaku Uto[1], Kazunori Sugiura[1], Shunsuke Maeda[2], Katsumasa Fujita[2], Takashi Washio[1], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ*, [2]*Grad Sch of Engin, Osaka Univ*)

Reversibly photoswitchable fluorescent proteins (RPFPs) are essential probes for highly-biocompatible super-resolution bioimaging with low phototoxicity. Although we recently developed a positively RPFP (pRPFP), Kohinoor, which was used for super-resolution microscopy such as SPoD-OnSPAN, nonlinear SIM, and RESOLFT, its fluorescence was not bright enough to take images of high spatial resolution and high S/N ratio. Here we show an improved pRPFP, which we derived by directed evolution from Kohinoor. The improved pRPFP, Kohinoor 2.0, exhibits 2.2-fold higher fluorescence and 5-fold faster maturation rate than Kohinoor. Thus, Kohinoor 2.0 is most likely to be useful for super-resolution imaging with higher spatial resolution in nonlinear SIM and SPoD-OnSPAN.

2Pos275 プラズモニックナノ粒子を用いた高速マルチカラー生体１分子イメージング
Multi-color and high-speed imaging of single biomolecules with plasmonic nanoparticles

Jun Ando[1,2], Akihiko Nakamura[1,2], Mayuko Yamamoto[1], Ryota Iino[1,2] ([1]*IMS, NINS*, [2]*SOKENDAI*)

Gold nanoparticles (AuNPs) have been used as optical probes to observe the fast motion of single biomolecules. Here, we developed a multi-color and high-speed single particle tracking system using silver and silver-gold alloy nanoparticles (AgNPs and AgAuNPs) together with AuNPs. We constructed total-internal reflection multi-color dark-field microscope with multiple lasers at 404, 473, and 561 nm that matches with plasmon resonance wavelength of AgNPs, AgAuNPs, and AuNPs, respectively. Spectrophotometer was used in the imaging optics, to project scattering image at each wavelength on the different portion of two-dimensional high-speed CMOS camera. Motions of the multiple lipids in membrane, and multiple kinesins were simultaneously observed at 100 μs time resolution.

2Pos276 Characterizing the spatio-temporal heterogeneity on biomolecular concentration, mobility and local environment in live cells

Sho Oasa[1], Aleksandar J. Krmpot[1,2], Stanko N. Nikolic[1,2], Lars Terenius[1], Rudolf Rigler[1,3], Vladana Vukojevic[1] ([1]*Dept. of Clin. Neurosci., Center for Mol. Med., Karolinska Inst.*, [2]*Inst. of Physics, Univ. of Belgrade*, [3]*Dept. of Med. Biochem. and Biophys., Karolinska Inst.*)

Cells convey key information via molecular interactions and transporting processes. They therefore precisely control concentration, mobility and immediate surroundings of biomolecules. To characterize in live cell the spatio-temporal dynamics of biomolecules and properties of their immediate surroundings, we have developed non-scanning quantitative confocal fluorescence microscopy with high spatio-temporal resolution (~240 nm, ~10 μs/frame) which relies on massively parallel Fluorescence Correlation Spectroscopy (mpFCS) to map the biomolecular concentration and mobility, and Fluorescence Lifetime Imaging Microscopy (FLIM) to map local immediate surroundings. We use here this method to map in live cells the dynamic landscape of transcription factor and opioid receptors.

2Pos277 Mapping of mechanical property on live cell surface by scanning ion conductance microscope

Satoko Kitazawa[1], Linhao Sun[2], Ayako Housaka[2], Takahiro Watanabe-Nakayama[2], Hiroki Konno[2], Mikihiro Shibata[2], Shinji Watanabe[2] ([1]*Grad. Sch. Math. &Phys., Kanazawa Univ.*, [2]*WPI-NanoLSI, Kanazawa Univ.*)

The mechanical properties of living cells are known to play crucial roles in many cellular processes such as motility, morphogenesis, and drug delivery. In this study, we will demonstrate the technique based on scanning ion conductance microscopy that can measure the spatial distribution of surface elastic responses of living cells with nanometer scale resolution. In addition, we will show results in the improvement of data throughput (pixels/sec) that is 10-100 times higher than that achieved with previous method.

2Pos278 定量的 ATP イメージングを用いた細胞の代謝状態の空間的相関の解析
Spatial correlation of metabolic states in mammalian cells revealed by quantitative single-cell ATP imaging

Hideyuki Yaginuma[1,2], Yasushi Okada[1,3] ([1]*BDR, Riken*, [2]*Grad. Sch. of Eng., Univ. of Tokyo*, [3]*Grad. Sch. of Sci., Univ. of Tokyo*)

Adenosine triphosphate (ATP) is a key molecule in metabolism that provides energy to intracellular reactions. Recently, we developed an ATP indicator protein “QUEEN-37C”, for visualization of ATP concentrations in mammalian cells at 37°C. The signal of QUEEN-37C agreed well with results from luciferase assay, and thus enabled us to measure and determine the single cell ATP. Using QUEEN-37C, we measured ATP concentrations in MDCK cell monolayer during metabolic inhibitor treatment. Interestingly, when cell density was high, some cells are more dependent on OXPHOS than other cells, and neighboring cells tended to show similar response to inhibitors. Our results suggest that neighboring cells are able to communicate so that they can adjust their metabolic states.

2Pos279 局所特徴モーフィングした補間による深さ解像度の低い 3 次元画像の改善
Feature based local morphing improved interpolation of 3D stack images at low depth resolution

Yutaka Ueno[1], Takashi Kawasaki[2], Totai Mitsuyama[1] ([1]*AIST Tokyo*, [2]*AIST Kansai*)

An algorithm to interpolate the cross section images for a 3D volume data was examined. In many case confocal microscopy for living cells provide depth stack images at less resolution interval than that of planer image. The 3D volume rendering and iso-surface rendering often resulted jaggy edges that should be smooth. Based on an assumption that imaging objects are larger than the depth resolution, we tested interpolating depth slice images between slice using feature based local morphing in which the pixel coordinate was deformed by simple local image alignment at small window size. Since results were satisfactory for sample medical MRI data, we are applying the method to the 3D volume data of medaka brain obtained by confocal microscopy.

2Pos280 スマートフォン発光顕微鏡による 1 細胞イメージング
Smartphone based chemiluminescence microscope for single cell imaging

Mitsuru Hattori[1], Sumito Shirane[2], Kuniaki Nagayama[2], Takeharu Nagai[1] ([1]*ISIR, Osaka Univ.*, [2]*Life is small. Company*)

There have been proposed some of smartphone-based methods to analyze biological phenomena. With a fluorescent tag, it is possible to detect a specific target in cells. However, the fluorescent emission requires a device for external light illumination. To mitigate this inconvenience, we introduced a chemiluminescence tag. Since the light emission doesn't require any light source, the size of device can be compact. An attachment lens to smartphone camera was newly developed to detect chemiluminescence from cells with high magnification with minimum optical aberration. By combining eNano-lantern previously developed by us, we succeeded single cell chemiluminescence imaging thorough the smartphone camera. This method could open a mobile imaging era.

2Pos281 鱗翅目昆虫の変態過程における極微弱バイオフォトン発光の連続画像計測
Continuous imaging of biophoton emission of lepidopterous insects during

Shoko Usui[1], Mika Tada[2], Masaki Kobayashi[1] ([1]*Grad. Sch. Elec., Tohtech*, [2]*Center for General Education., Tohtech*)

Spontaneous ultra-weak photon emission from living organisms, referred to as biophoton emission, is a generally observed phenomenon irrespective of the organism species, which is attributed to the production of excited molecules in a metabolic reaction, especially involving reactive oxygen species. Application of biophoton to extract physiological information has been widely studied using plants and mammals. However, biophoton properties of insects remain unclear. In this study, we have detected biophoton images of holometabolous insects, Papilio protenor, during pupation as a moving picture using our developed system and clarified spatiotemporal characteristics of biophoton emission, representing physiological changes underlying the mechanism of morphological changes.

2Pos282 Construction of a millisecond structured illumination microscope and its application to ultrafast super-resolution live cell imaging

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We constructed a millisecond structured illumination microscope (SIM) for the observation of dynamical changes inside living cells with super-resolution (SR). To achieve the fast SIM imaging, we introduced two Pockels cells into the SIM, which enables us to shift the phase of structured illumination patterns at high speed (up to 20 kHz). By using a high-speed camera synchronized with the Pockels cells, we acquired five images consecutively with a structured illumination pattern having different phase shifts within each 1 ms, and obtained a single SR image using the 5 images. As the first demonstration of fast SR live cell imaging, we obtained a series of SR images of living cells stained by fluorescence nanoparticles with spatio-temporal resolution of ≦150 nm and 1 ms.

2Pos283* DNA を用いた反応拡散系のプログラムによるハイドロゲル中におけるパターン形成
Programmable reaction-diffusion system using synthetic DNA for pattern formation in hydrogel medium

Keita Abe, Ibuki Kawamata, Shin-ichiro Nomura M., Satoshi Murata (*1Department of Robotics, Graduate School of Engineering, Tohoku University, Japan*)

Multicellar organisms have the capability to develop complex and functional shapes. In the process of the development, morphogenesis occurs with pattern formation by biological phenomena such as reaction and diffusion of biomolecules. The biological development has inspired us to engineer self-organizing system. By designing reaction and diffusion, we can fabricate a system which forms desired pattern automatically. Here, we demonstrate a method of pattern formation based on an artificial reaction diffusion system in hydrogel medium using synthetic DNA. Using the method, we successfully demonstrated weighted Voronoi pattern formation with changing the weight by tuning DNA condition. We will report the method of programmable pattern formation and its latest result.

2Pos284* マイクロドロップレットを用いた G タンパク質共役型受容体ペプチドアゴニスト探索法のフィジビリティスタディ
Feasibility study of the method to obtain peptide agonists for G protein-coupled receptors using water-in-oil microdroplets

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G protein-coupled receptors (GPCRs) are one of the most important drug-targets in the pharmaceutical industry. We developed a method to obtain peptide agonists for GPCRs, which is a combination of in vitro selection with in vitro compartmentalization and a yeast-based fluorescent reporter assay for GPCR signaling. To confirm the feasibility of the method, pilot experiments were performed with somatostatin (SST) and its receptor (SSTR2). DNA constructs encoding wild-type and mutant SST were mixed at a ratio of 1:100 and 1:1,000 to prepare a model library. We successfully recovered the sequence encoding wild-type SST from the library. Now, we are trying to obtain the wild-type SST sequence from a library of partially randomized SST sequences.

2Pos285* 高分子分解酵素産生微生物の取得のための液滴の変形能を利用したスクリーニング法
Deformability-based microfluidic droplet screening to obtain microbes producing macromolecule-degrading enzymes

Mikihisa Muta[1], Kai Saito[1], Ryo Iizuka[1], Wataru Kawakubo[2], Dong Hyun Yoon[3], Tetsushi Sekiguchi[3], Shuichi Shoji[3], Mei Ito[4], Yuji Hatada[4], Takashi Funatsu[1] ([1]*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*, [2]*Dept. of Nanosci. and Nanoeng., Waseda Univ.*, [3]*Res. Org. for Nano & Life Innov.*, [4]*Dept. of Life Sci. and Green Chem., Saitama Inst. of Technol.*)

Environmental microbes secrete enzymes capable of degrading macromolecules into smaller molecules that can be used as nutrients for growth. The enzymes have potential benefits for effective utilization of terrestrial resources such as biomass. We devised a deformability-based microfluidic droplet screening method to obtain microbes producing macromolecule-degrading enzymes. Microbes are encapsulated in water-in-oil (W/O) droplets at the single-cell level. The W/O droplets, where the macromolecules are progressively degraded into smaller molecules, increase the deformability. The screening is achieved by using a microfluidic device that enables passive sorting of deformable W/O droplets. Using this method, we have demonstrated agarose-degrading microbes can be obtained.

2Pos286 カップ形状微小電極の開発と 1 細胞発現分子計測への応用
Development of cup-shaped microelectrode and its application for detection of expressed biomolecules in single cell level

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We propose a new method to detect biomolecules expressed on a cell surface based on the measurement of electrochemiluminescence (ECL). For the achievement, a cup-shaped microelectrode, whose inner concave was composed of nano-carbon layer and the outer was nickel, was fabricated. Target molecules (EpCAM in this study) on cell surfaces were labeled with the antibody conjugated with ruthenium complex, an ECL substrate, and the labeled cells were trapped to inner concave of the cup-shaped microelectrode. When electric voltage was applied, ECL was observed from the cell-captured cups. By using this method, cancer cells on which target molecules were a few expressed can be successfully detected, indicating quite high target detection sensitivity of the developed method.

2Pos287 太陽光に依存しない細胞内生物発光による光合成
Sunlight independent plant cell photosynthesis by self-contained bioluminescence

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Plants use external light sources such as sunlight for photosynthesis, which fixes CO2 and generates O2, but the use of internal bioluminescence as an energy source has not been explored. To determine whether bioluminescence emitted within a plant could be used as a light source for photosynthesis, we generated transgenic liverworts, Marchantia polymorpha, expressing green-, or red-enhanced Nano-lanterns (eNLs) emitting visible intensities of light upon addition of substrate. Liverwort expressing red-eNL produced O2, which was prevented by photosystem II (PSII) inhibitors, and consumed CO2 via photosynthesis. This glowing plant provides a useful platform for future technologies including solar energy-independent photosynthetic devices.

2Pos288 (2SEP-7) グラフェン電界効果トランジスタとフェムトリットルチャンバーを用いたデバイ遮蔽を超える電気的バイオセンシング
(2SEP-7) Electrical Biosensing beyond the Debye Screening Length Using Graphene Field-Effect Transistor in Femtoliter Microchamber

Takao Ono[1], Yasushi Kanai[1], Koichi Inoue[1], Yohei Watanabe[2], Shin-ichi Nakakita[3], Toshio Kawahara[4], Yasuo Suzuki[4], Kazuhiko Matsumoto[1] ([1]*ISIR, Osaka Univ.*, [2]*Kyoto Pref. Univ of Med.*, [3]*Kagawa Univ.*, [4]*Chubu Univ.*)

Graphene is 2D material which has the highest carrier mobility in known materials. Graphene field-effect transistor (G-FET) has a potential for electrical biosensing, because it transduces the carrier modulation by charged targets to high conductivity change. However, in physiological ionic strength, electrical field around the target is attenuated within 1 nm, due to Debye screening. Here we show an electrical biosensing utilizing enzymatic products of the target, which diffuse and reach the graphene surface. 1) Encapsulated into microdroplet to accumulate the products, G-FET monitored ammonia production of urease in real time. *Helocobacter pylori* was detected owing to its urease reaction in sensitivity below single bacterium. 1) T. Ono *et al., Nano Lett., accepted*.

3Pos001 Dynamic docking between a flexible enzyme and its inhibitor using multicanonical MD simulations and binding free energy calculations

Narutoshi Kamiya[1], Gert-Jan Bekker[2] ([1]*Sim. Stu., Univ. Hyogo*, [2]*IPR, Osaka Univ.*)

We have performed dynamic docking between a flexible receptor with a wide binding pocket and a highly flexible, medium sized drug by employing McMD simulations. We applied our method to predict the native binding configuration and sample the intermediary binding structures between the enzyme β-secretase 1 and its inhibitor. Representative structures located at free energy minima obtained from McMD were taken and subjected to canonical MD to refine and validate, reproducing the experimental native complex structure. The binding free energy was estimated by umbrella sampling along representative pathways obtained from the McMD ensemble, followed by weighted histogram analysis to estimate the affinity, which also reproduced the experimental inhibitory affinity.

3Pos002 Biological Structure Model Archive: 計算機で得られた生体分子モデルのアーカイブ
Biological Structure Model Archive: An archive for computationally obtained data

Gert-Jan Bekker, Takeshi Kawabata, Genji Kurisu (*Osaka University, IPR*)

We present BSMA, which aims to collect data obtained via in-silico methods related to structural biophysics. Publishing their data enables researches to not only have an external backup, but also provide an opportunity to promote their work via an interactive platform and providing third-party researchers access to the raw data. Besides uploading the raw data, BSMA enables users to annotate the data with additional explanations and figures. Furthermore, via our WebGL based molecular viewer Molmil it is possible to recreate 3D scenes as shown in the corresponding scientific article in an interactive manner. The archive is available at https://bsma.pdbj.org, where published entries can be freely downloaded, while new entries can be submitted using users' ORCID ID.

3Pos003 Nanodisc に再構成した好熱菌由来 V-ATPase の単粒子解析
Single-particle analysis of the lipid nanodisc-reconstituted V-type ATPase/synthase from *Thermus thermophilus*

Atsuko Nakanishi[1], Jun-ichi Kishikawa[1], Kaoru Mitsuoka[2], Ken Yokoyama[1] ([1]*Faculty of Life Sci. Kyoto Sangyo Univ.*, [2]*Res. Ctr. for UHVEM. Osaka Univ.*)

V-ATPase is a rotary molecular motor which couples ATP hydrolysis/synthesis in V_1 with proton flow in V_o through rotation of central rotor apparatus. Here, we determined the structure of lipid nanodisc-reconstituted V-ATPase from *Thermus thermophilus* (*Tth* V-ATPase) by single-particle analysis using cryo-electron microscopy (cryo-EM). The use of nanodiscs allowed to hold *Tth* V-ATPase in soluble form without detergents, resulting in a higher resolution map than our previous data. The local resolution of V_1 was sufficient to construct the near atomic model, while densities were not well resolved in V_o. We are attempting to determine the structure by using a large dataset to classify more homogeneous particles and build an atomic model of the V-ATPase.

3Pos004 Prediction of ligand distribution around a protein by 3D-RISM theory

Masataka Hamano, Masatake Sugita, Takeshi Kikuchi, Fumio Hirata (*Dept. Bioinfo., Coll. Biosci., Ritsumeikan Univ*)

The three-dimensional reference interaction site model (3D-RISM) theory which is a statistical mechanics theory of molecular liquids can analytically predict the distribution of the solvent inside and around a solute in solution. Solving 3D-RISM theory, it is possible to determine the distribution of a ligand inside and around the protein by mixing ligand molecule in the solvent. However, it has not been clarified how accurately the binding position of a ligand fragment can be predicted by using this method. Then, for the purpose to examine the accuracy of this method, we apply to some protein-ligand systems, T4 Lysozyme-benzene, IGG-KAPPA protein-isopropanol and SH3 domain-imidazole.

3Pos005 アミノ酸配列情報からのフラボヘモグロビンのフォールディング機構予測
Prediction of the folding mechanism of flavohemoglobin based on average distance statitistical method

Maho Osugi, Takeshi Kikuchi (*Dept. Bioinfo., Coll. Biosci., Ritsumeikan Univ.*)

In this study, we predict their folding mechanism of flavohemoglobin a chimeric protein of myoglobin and reductase by mean of sequence analysis. Average distance map (ADM) is used in this study to predict the location where a structural compact region is formed in an initial state of folding. The results are compared with the 3D structure of the homologous proteins of which structures are known. As a result of ADM, four predicted compact regions are predicted. A Rossmann fold at C-terminus of flavodoxin is expected to fold first, due to the highest η-value. Next, the globin domain starts fold at the N-terminus, followed by folding of the β-barrel at the center of flavodoxin protein.

3Pos006 分布推定アルゴリズムによる単粒子解析投影パラメーター決定
Determination of projection parameters in single particle analysis using Estimation of Distribution Algorithms

Nobuya Mamizu[1,2], Takuo Yasunaga[1] ([1]*Kyushu Institute of Technology*, [2]*SYSTEM IN FRONTIER INC.*)

We propose a method for determining each projection parameter of particle images in single particle analysis. We apply Estimation of Distribution Algorithms (EDAs) to calculate prior probability distribution in the reconstruction model based on RELION. This makes it possible to adaptively change parameter search ranges and density from global to local without dividing the parameter space as RELION. Using simulation 2D images generated from a known 3D structure, the method was evaluated for the cases with and without a initial model. In both cases, the projection parameters of each particle converged correctly, and we reconstructed the original 3D structure. We will evaluate this method for actual electron microscope images.

3Pos007 連続滴定小角 X 線散乱測定を用いた KaiC に対する KaiA の滴定挙動解析
Titration analysis of KaiA for KaiC using continuous titration small angle X-ray scattering

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In the cyanobacterial circadian clock, assembly and disassembly of Kai proteins contribute to robust phosphorylation cycle of KaiC. To investigate the binding behavior of KaiA and KaiC with different phosphorylation levels, we carried out continuous titration SAXS experiments using μ-fluidics based auto sampling system. Both of phosphorylated (p-) and dephosphorylated (dp-) KaiC shows an obvious increase in the cross term reflecting KaiA/C complex formation. While the titration curve of dp-KaiC can be represented by the simple binding of KaiA2 and KaiC6, that of p-KaiC implies multiple binding modes, suggesting that KaiC possesses several binding ways of KaiA depending on the phosphorylation level.

3Pos008 Folding properties prediction of ribonuclease and chymotrypsin based on inter-residue average distance statistics

K M Ahsanul Kabir, Takeshi Kikuchi (*Dept. Bioinfo., Grad. Sch. of Life Sci., Ritsumeinkan Univ., Computational Biomolecular Chemistry lab.*)

Protein folding is exceedingly complex process that produce unique 3D structure of proteins. Decoding the folding information from the amino acid sequences of a protein is a changeling task in computational biology. According to SCOPe classification two different class protein ribonuclease and chymotrypsin are investigated in this study. We have been applying a contact map and a contact frequency prediction technique to evaluate the protein folding. Finally determine the evolutionary conserved hydrophobic residues and hydrophobic packing. The initial folding of those protein starts in their N-terminal and then the C-terminal is formed which stabilize the native structure of both of this class protein.

3Pos009 The off-axis rotor of *Enterococcus hirae* V-type ATPase by Volta phase contrast cryo-EM

Raymond N. Burton-Smith[1], Jun Tsunoda[1], Yu Yamamori[2], Naoyuki Miyazaki[3], Fabiana L. Imai[4], Chihong Song[1], Kentaro Tomii[2], Kenji Iwasaki[3], Junichi Takagi[5], Hiroshi Ueno[7], Takeshi Murata[4], Ryota Iino[6], Kazuyoshi Murata[1] ([1]*NIPS*, [2]*AIST*, [3]*Univ. Tsukuba*, [4]*Chiba Univ.*, [5]*Osaka Univ.*, [6]*IMS*, [7]*Univ. Tokyo*)

The V-type ATPases are large membrane complexes which utilise ATP hydrolysis to power ion transport across cell membranes, in this case the alkali metal ions Li+ and Na+. Our previous report demonstrates displacement of the rotor axis in the Enterococcus hirae V-ATPase. *E. hirae* is a Gram-positive zoonotic bacterium, with implications for both human health and the farming industry. Here we have extended the previous report using cryo-EM data acquired on a Titan Krios equipped with a Falcon III direct detector and Volta-type phase plate. In a detergent-solubilised complex, a single conformation dominates, while the complex as a whole also shows significant orientation preference. Utilising a PA-tag system, we isolated a second conformation of the V-ATPase complex.

3Pos010 GTP 結合型および GDP 結合型微小管におけるチューブリン C 末端構造分布差の分子動力学計算による解析
Simulation study for conformational difference of tubulin C-terminal tails in GTP-bound and in GDP-bound microtubule

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The nucleotide state of tubulin is important for microtubule stability. The microtubule composed of GTP-bound tubulin is stable, while those of GDP-bound tubulin is unstable, leading to collapse. The nucleotide state may affect conformations of C-terminal tails (CTTs) of tubulin. CTTs are necessary for microtubule to form native cylindrical assembly. However, the relationship between CTT conformation and microtubule stability remains unclear. Then, we conducted molecular dynamics simulation for microtubule systems (3x3 tubulin heterodimers) binding of GDP or GTP. We found that CTTs at interdimer are distributed close to each other in GDP-form, suggesting that electrostatic repulsion between the negatively charged CTTs might play a role in heterodimer disassembly.

3Pos011 距離依存誘電率および溶媒接触表面積を取り込んだ SAAP 力場を用いたトリプケージの分子シミュレーション
Molecular simulation of Trp-cage using the SAAP force field with Distance-Dependent Dielectric and Solvent Accessible Surface Area

Koji Yoshida, Taku Shimosato, **Michio Iwaoka** (*Tokai Univ., Depart. Chem.*)

We recently performed replica-exchange MC simulation for chignolin (GYDPETGTWG) using SAAP, a new force field developed in our laboratory, and obtained the native structure (all-atom RMSD = 1.5 Å) in 8.5 % [1]. However, for Trp-cage (NLYIQWLKDGGPSSGRPPPS), the native-like structure with a large main-chain RMSD of 4.4 Å was only obtained. To incorporate solvent effects more adequately, we herein introduced two additional energy terms of Distance-Dependent Dielectric (DDD) and Solvent Accessible Surface Area (SASA) into the SAAP. By introducing DDD the native-like structure with a smaller RMSD of 3.7 Å was mainly obtained for Trp-cage, but introduction of both DDD and SASA did not improve the accuracy. [1] M. Iwaoka, et al., J. Comput. Aided Mol. Design, 2017, 31, 1039.

3Pos012 asymmetry of psi-loop motifs

Koki Fukuda, George Chikenji (*Dept. Appl. Phys., Grad. Sch. Eng., Nagoya Univ.*)

The psi-loop motifs are defined by consecutive three beta strands on the amino acid sequence that include hairpin connection with one intervening strand, and they have four possible distinct connecting topologies. Although these four psi-loop topologies are structurally similar to each other, the frequencies of these topologies in the protein structure database are largely different: only the two of them are observed frequently and the others are rare. Addressing a question why their observed frequencies are so different is important for protein structure prediction and design, but a consensus has not been reached. Here we study the origin of the difference by the database analysis and physics based simulations.

3Pos013 溶液 NMR による水素結合の直接観測
Direct observation of hydrogen bonds by solution NMR

Taiki Koizumi, Hiroki Nakajima, Yutaka Ito, Masaki Mishima (*Grad. Sch. Sci., TMU*)

Hydrogen bonds play an important role in structures of biological molecules and expression of their functions. X-ray crystallography, one of the major structural analysis methods analyzing electron density, is not suitable for directly observation of hydrogen atoms. Meanwhile, hydrogen atoms can be observed directly in NMR, and hydrogen bonds can be detected directly by observing the J couplings via hydrogen bonds which caused by a fermi contact term derived from 1S orbital electron of hydrogen atom. In this study, we used Bacillus thermoproteolyticus Ferredoxin (BtFd) as a target protein and attempt to directly observe bifurcated hydrogen bonds, which are suggested in X-ray crystal structure analysis, using long-range HNCO measurements.

3Pos014 (3SHA-5) STAP-2 により Breast tumor kinase が活性化する機構の解明
(3SHA-5) Molecular basis of Breast tumor kinase by an adaptor protein, STAP-2

Junki Nakasako[1], Yuki Matsuo[2], Ryo Kanda[2], Yoshino Tanaka[2], Min Yao[3], Tadashi Matsuda[2], Katsumi Maenaka[2], Toyoyuki Ose[2,3,4] ([1]*Graduate school of Life Science*, [2]*Faculty of Pharm.*, [3]*Faculty of Advanced Life Science, Hokkaido University*, [4]*JST PRESTO*)

Breast tumor kinase (Brk) is a nonreceptor tyrosine kinase present in cytosol. Although Brk is expressed frequently in breast cancer cells, usually, it is not observed in normal breast tissue, therefore the possibility of therapeutic application targeting Brk has been pointed out. It has been reported that Brk over-phosphorylates the transcription factors STAT3 and STAT5 through the interacting with signal-transducing adaptor protein-2 (STAP-2) thus causes abnormal cell proliferation. We believe it is important to understand the activation mechanism of Brk. To identify important domains for the activation of Brk, we constructed recombinant expression system of STAT-2 including some mutants. The interaction analysis and enzymatic assay are now now in progress.

3Pos015 分子シミュレーションで探る angiotensin II type 1 receptor（AT1R）の活性化メカニズム
Activation mechanism of the angiotensin II type 1 receptor (AT1R) explored by molecular simulations

Yuichiro Kanamori, **Tadaomi Furuta**, Minoru Sakurai (*Center for Biol. Res. & Inform., Tokyo Tech*)

Angiotensin II type 1 receptor (AT1R) belongs to the G protein-coupled receptor (GPCR) superfamily, composed of seven transmembrane helices, and mediates diverse pathological actions in cardiovascular, renal, and so on. Although it has been revealed that AT1R is activated by Angiotensin II or mechanical stress, the detailed mechanism is still unclear. Moreover, several constitutively active mutants (CAMs) are known so far. In this work, to elucidate the activation mechanism of AT1R, we investigated the wild type and a CAM of AT1R by molecular simulations. As a result, it was found that fluctuation of the middle region of TM6 around the toggle switch was important for the activation, which was related to the weakening of the hydrophobic interaction between TM6 and TM3.

3Pos016 クライオ電子顕微鏡による多剤排出ポンプ複合体 MexAB–OprM の構造解析
The wild-type structures of MexAB-OprM multidrug efflux pump revealed by cryo-electron microscopy

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In Pseudomonas aeruginosa, MexAB-OprM plays a central role in multidrug resistance by ejecting various antibiotics. In this presentation, we present the structures of wild-type MexAB-OprM in the presence or absence of drugs at near-atomic resolution by cryo-electron microscopy single-particle analysis. The structure reveals that OprM does not interact with MexB directly, and that it opens its periplasmic gate by forming a complex. Furthermore, we confirmed the residues essential for complex formation with the in vitro and in vivo functional analyses, and observed a novel movement of the drug entrance gate. Based on these results, we propose mechanisms for complex formation and drug efflux.

3Pos017 IgG 抗体の Fv フラグメントはドメイン運動に伴う水和構造変化によって抗原認識ループの構造変化を制御する
Domain motion of Fv-fragment in antibody immunoglobulin G controls conformation of antigen-recognizing loop through hydration structure

Tomotaka Oroguchi[1,2], Masayoshi Nakasako[1,2] ([1]*Faclt. Sci. Tech., Keio Univ.*, [2]*RIKEN SPring-8 Center*)

Immunoglobulin G is a representative member of antibodies, and recognized antigens by the Fv-fragment in the Fab portion. The Fv-fragment is composed of VH and VL domains, and a flexible loop in VH domain functions as an antigen-recognizing region (CDRH3 region). The previous structural studies have indicated that CDRH3 region undergoes conformational transition between inhibitory and active states, and the transition correlates with domain rearrangement of the two domains. In this study, we investigated conformational dynamics of Fv fragment by using all-atom molecular dynamics simulations. The results demonstrates that the domain motion of VL domain against VH domain controls the conformational transition of CDRH3 region through changes in hydration structure.

3Pos018 3D–RISM 理論を応用した溶液中における Met–enkephalin の構造揺らぎの解析
Analysis of structural fluctuations of Met-enkephalin in the solution phase by means of 3D-RISM theory

Masatake Sugita[1], Fumio Hirata[2] ([1]*Dept. of Bioinfo., Col. of Life Sci., Ritsumeikan Univ.*, [2]*Toyota Phys. & Chem. Res. Inst.*)

Recently, we have tried to implement an analytical theory for estimating fluctuations of a solute molecule in solution. In this formulation, variance-covariance matrix is expressed as an inverse matrix of the hessian matrix of the free energy surface that can be defined by the sum of the potential energy and solvation free energy of the protein. In this study, we analyze structural fluctuation of Met-enkephalin immersed in water by calculating the second order derivative of the solvation free energy in addition with the potential energy, and diagonalizing the hessian matrix. After that comparing the results with those from the Normal Mode analysis and MD simulation.

3Pos019 網羅的構造解析による酵素機能関連天然変性蛋白質の探索
Exploration of disordered proteins associated with enzymatic functions by comprehensive structural search

Satoshi Omori[1], Hafumi Nishi[1], Kengo Kinoshita[1,2] ([1]*GSIS, Tohoku Univ.*, [2]*ToMMo, Tohoku Univ.*)

Intrinsically disordered regions (IDR) are protein regions that lack stable 3D structures, and yet they contribute to biological functions. Although enzymatic reactions are assumed to be achieved via stable structures of the enzymes, they often involve disorder-to-order (or vice versa) transitions of IDRs. In this study, we explored functionally associated IDRs using Natural Ligand Database, a structural database of enzyme-ligand complexes. The functional associations were estimated by statistical significance, and the influences of crystal contacts were eliminated. We identified 196 enzymes with functional IDRs, and in some cases, the association between IDRs and known molecular mechanisms was confirmed. New functional IDRs are also expected to be detected.

3Pos020 A local structural environment descriptor toward evaluating impact of rare variants in humans on protein structures and functions

Atsushi Hijikata, Masafumi Shionyu, Tsuyoshi Shirai (*Nagahama Inst. Bio-Sci. Tech.*)

Although recent advances in genome sequencing technologies make it easy to identify genomic variants in individuals, it remains a challenge to interpret the impact of a number of variants of uncertain significance (VUS). Clarifying the functional significance of these VUSs is expected to lead to uncover the genotype-phenotype connections. To this end, we introduced a local structural environment descriptor (LSED), which is a set of amino acid residues represented as vectors from Cα to Cβ atoms surrounding a given residue, to evaluate the impact of VUSs. LSEDs for 10,217 pathogenic mutations in 2,655 proteins of known 3D were quantitatively compared each other. We found structural similarities between LSEDs from non-homologous proteins in 15% of the pathogenic mutations.

3Pos021 A new MD integration enabling large time step from accurate temperature and pressure evaluations

Jaewoon Jung[1,2], Hiraku Oshima[3], Kento Kasahara[3], Chigusa Kobayashi[1], Takaharu Mori[2], Yuji Sugita[1,2,3] ([1]*RIKEN Center for Computational Science*, [2]*RIKEN Cluster for Pioneering Research*, [3]*RIKEN Center for Biosystems Dynamics Research*)

We present a new MD integration method that enables a large time step and implement it in GENESIS MD software. It is based on the accurate evaluation of temperature and pressure combined with hydrogen mass repartitioning (HMR) scheme. It also includes multiple time step integration (MTS) based on group approach for temperature and pressure evaluations. The new integration is tested extensively by investigating structural/physical properties of lipids, peptides, proteins, and nucleic acids in multiple microsecond MD simulations. Our new integration is shown to be reliable up to 5 fs time step for structure, dynamics and free-energy differences of all the test molecules. This method allows more than twice speed-up of the conventional and enhanced-sampling MD simulations.

3Pos022 Protein interactions in the *in vitro* cyanobacterial circadian clock system revealed by SDSL-ESR

Risa Mutoh[1], Takahiro Iida[1], Hiroyuki Mino[2], Masahiro Ishiura[3] ([1]*Faculty of Sci, Fukuoka Univ.*, [2]*Dep. Sch. of Sci., Nagoya Univ.*, [3]*CGR, Nagoya Univ.*)

Cyanobacterial circadian clock machinery is composed of KaiA, KaiB and KaiC proteins. The three proteins interact each other, and the size and stoichiometry of Kai protein complex change during circadian cycle. We previously revealed that the KaiB, labeled specifically at 64th residues with the spin label MTSSL, directly interacted with KaiA C-terminal domain by electron spin resonance analysis. Further, we measured 15 Cys-substituted mutants of KaiB in the presence of KaiA. We detected the ESR spectral changes in several KaiB mutants, and mapped the KaiA interacting sites on KaiB. These were located inside of KaiB molecules, suggesting that the conformation of KaiB dramatically changed in the presence of KaiA.

3Pos023 Investigation of formation mechanism of Prx high molecular weight complexes

Mami Jindai[1], Rino Sasaki[1], Noriyuki Kodera[2], Toshio Ando[2], **Hiroki Konno**[2] ([1]*Sch. of Nat. Syst., Coll. of Sci. & Eng., Kanazawa Univ.*, [2]*WPI Nano Life Sci. Inst. (WPI-NanoLSI), Kanazawa Univ.*)

The function of ubiquitous 2-Cys peroxiredoxins (Prxs) can be converted alternatively from peroxidases to molecular chaperones. This conversion has been reported to occur by the formation of high molecular weight (HMW) complexes upon overoxidation of or ATP/ADP binding to Prxs. We first demonstrated that negatively charged lipids are essential for functional and structural switch of human Prx II. However, the formation mechanism and physiological significance of the Prx HMW complex containing lipids still remain unclear. To reveal the formation mechanism of Prx HMW complex with lipids, we tried to visualize the formation process of Prx HMW complex using high-speed AFM. We also identified the lipid binding site on Prx by site-directed mutagenesis analysis.

3Pos024 Time-localised Predictions of Conformational Transitions in Protein Dynamics

Ryuhei Harada[1], Vladimir Sladek[2], Yasuteru Shigeta[1] ([1]*CCS, Unv. of Tsukuba*, [2]*Slovak Academy of Sciences*)

In this study, we introduces a new descriptor for identifying conformational transitions in molecular dynamics (MD) simulations of proteins. We also adopt the descriptor in an enhanced conformational sampling protocol known as nt-PaCS-MD. The new descriptor, moving root-mean-square deviation (mRMSD), facilitates the analysis of a series of short MD simulations to identify perturbed structures. In nt-PaCS-MD, mRMSD is used as a measure to select starting geometries of short MD simulations. By demonstrations, mRMSD apparently improved the effectiveness of the nt-PaCS-MD protocol. Additionally, we successfully constructed free energy landscapes based on Markov state models built from short trajectories of nt-PaCS-MD capturing transitions between different states.

3Pos025 フレキシブルドッキングによる結合自由エネルギーと速度定数計算
Calculation of binding free energy and kinetic rates with flexible protein docking

Duy Tran, Akio Kitao (*Tokodai, Grad. Life Sci. Tech.*)

Molecular association and dissociation are among the important processes of living organism. In this presentation, we will report our recent developed method for generating dissociation pathways achieved by the combination of PaCS-MD and the Markov state model. The method is able to calculate the binding free energy and the kinetic rates of a protein/flexible peptide complex in agreement with experiments. As an extension of the above method, we developed a flexible docking procedure, which can predict the complex structure of a protein and a flexible peptide using the ranking of the docking-generated decoys with binding free energy. Our new method offers the abilities of predicting the complex structure and calculating binding free energy and kinetic rates.

3Pos026 Evolutionary diversity of Kai-protein clock system in cyanobacteria

Atsushi Mukaiyama[1,2], Dongyan Ouyang[1], Yoshihiko Furuike[1,2], **Shuji Akiyama**[1,2] ([1]*IMS*, [2]*SOKENDAI*)

KaiC from *Synechococcus elongatus* PCC 7942 is a multifunctional enzyme with autokinase, autophosphatase, and ATPase and these enzymatic activities show a rhythm with a period of ~24 h in the presence of KaiA and KaiB. Although an interplay among the three functions is required as the core of the clock, it remains little unknown whether and how KaiC from other cyanobacterial species retains these properties. To address this, we picked up various KaiC homologues on the basis of a phylogenetic tree and examined their biochemical functions. So far, we prepared ~10 KaiC homologues and found that some species show a circadian rhythm by co-incubating with KaiA and KaiB. We will discuss evolutionary diversity of the cyanobacterial clock system.

3Pos027 Hierarchical classification method of protein-protein interfaces based on their secondary structures

Takashi Fujii, Kazuo Fujiwara, Masamichi Ikeguchi (*Grad. Sch. of Eng., Soka Univ*)

Protein-protein interactions play crucial roles in many biological functions. It is also known that structurally unrelated proteins have occasionally similar interfaces. The purpose of this study is to develop a new method to evaluate similarity between two interfaces based on their secondary structures and construct hierarchical classification. We extracted 7,671 PDB entries of homodimer with a single domain. First, the features of the interfaces of homologous proteins were compared to examine their diversity. Then, we developed a similarity evaluation method applicable to a pair of complexes having different structures. Next, we constructed a hierarchical classification of dataset entries based on this method.

3Pos028 タンパク質ダイナミクスに対する拡散マップ法の適用
Applications of a diffusion map method to protein dynamics

Hiroto Kikuchi[1], Ayori Mitsutake[2], Hiroshi Fujisaki[1] ([1]*Dept. of Phys. Nippon Med. Sch.*, [2]*Dept. of Phys. Meiji Univ.*)

Principle component analysis (PCA) is a widely used method to extract functionally relevant and low dimensional motions in proteins or peptides but has some limitations. Relaxation mode analysis (RMA), on the other hand, is a new technique to extract slowest collective motions in proteins or polymer systems, and in some cases can capture important collective motions which might be overlooked by PCA. Recently we have found that some collective motions in a small peptide, chignolin, calculated by another manifold learning technique, diffusion map, correlate very well with those calculated by RMA. We further compare the two methods by applying them to other proteins systems, protein-G, PIN1 enzyme, and so on and discuss the similarity and difference.

3Pos029 myPresto, computer-aided drug development software

Shinji Iida[1,11], Ikuo Fukuda[2], Junichi Higo[2], Kota Kasahara[3], Takashi Kurosawa[4], Tadaaki Mashimo[5], Kiyotaka Misoo[5], Yoshinori Wakabayasi[6], Ryuta Murakami[1,7], Chisato Kanai[1,7], Yusuke Sugihara[8], Mitsuhito Wada[1], Hironori Nakamura[9], Yoshifumi Fukunishi[10] ([1]*NNPC*, [2]*Grad. Sch. Sim. Hyogo Univ.*, [3]*Col. of Life Sci., Ritsumeikan Univ.*, [4]*Hitachi Solutions East Japan, Ltd.*, [5]*IMSBIO Co., Ltd.*, [6]*BY-HEX LLP*, [7]*INTAGE Healthcare Inc. Drug Discovery Suport Department*, [8]*Fujitsu Kusyu Systems Ltd.*, [9]*Biomodeling Research Co., Ltd.*, [10]*AIST, molprof*, [11]*JBiC*)

myPresto is an open source software for drug development and has been developed since 2001. The software implements a variety of computational methods: molecular dynamics (MD) simulation, molecular docking simulation, virtual compound screening, etc, and therefore all-inclusive for computer-aided drug design. Here, we demonstrate how myPresto works, exhibiting its effectiveness: (1) myPresto integrated with a graphical user interface (myPresto portal), (2) Multi-batch PCR to predict molecular properties (solubility, membrane permeability, distribution coefficient), (3) a docking-score QSAR method to estimate binding-free energy, and (4) an application of an enhance sampling MD simulation to protein-protein interactions.

3Pos030 十二量体フェリチン様 Dps の希土類バイオミラリゼーションにおける構造基盤
Structural basis of rear-earth metal biomineralization in dodecameric ferritin-like protein, Dps

Mitsuhiro Okuda[1,2], Pretre Gabriela[1], Kornelius Zeth[3] ([1]*CIC nanoGUNE*, [2]*Ikerbasque*, [3]*Roskilde University*)

Dps (DNA-binding protein from starved cells) forms a dodecameric cage-shaped protein complex (9 nm diameter) for iron uptake and storage, leading to inhibition of the Fenton reaction. The protein complex with a molecular weight of ~240 kDa has a cavity at the center of the protein complex (4.5 nm diameter) providing storage space for up to 500 iron atoms for biomineralization. In this work, we analyzed the structures of Listeria innocua Dps including trivalent ions (La, Tb), rear-earth metal using synchrotron radiation. At the ion channels and ferroxidase centers, the number of the rear-earth metals ions varied depending on ion species along with significant conformational changes of aspartic and glutamic acids side chains relative to the apo-protein without metal ions.

3Pos031 演題取り消し

3Pos032 天然構造が極めて類似したタンパク質の熱変性：遷移構造が異なる分子起源の理論研究
A theoretical study for thermal unfolding of proteins with quite similar native structure and different transition structures

Ken Tomihara, Takashi Yoshidome (*Department of Applied Physics, Tohoku University*)

Goat alpha-lactalbumin and human lysozyme share almost the same tertiary structure but different transition structures. To elucidate the molecular origin of the difference, we performed molecular dynamics simulations of unfolding of these proteins. To analyze the trajectories, both proteins were decomposed into seven segments in accordance with the previous study [1]. After classifying the trajectories into the native, unfolded, and in-between states, the Q values of segments and the interfaces between segments were computed. In the presentation, we will discuss molecular origin of the difference in the transition structures through comparing the Q values. [1] T. Oroguchi, M. Ikeguchi, M. Ohta, K. Kuwajima, and A. Kidera, J. Mol. Biol., 371, 1354 (2007).

3Pos033 分子動力学法と機械学習を用いたテトラペプチドの凝集性の評価
Evaluating aggregation propensity of tetra-peptide using MD and machine learning

Yoichi Kurumida, Yutaka Saito, Tomoshi Kameda (*AIRC, AIST*)

Evaluation of the aggregation propensity of a peptide is important to elucidate protein property. It is also useful in protein engineering for avoiding the aggregation of designed proteins. It has been reported that di-peptides and tri-peptides aggregation propensity can be evaluated using molecular dynamics (MD) simulation. However, such methods are difficult to be applied to tetra-peptides due to a large sequence space (160,000 types). In this study, we combined MD simulation and machine learning to accelerate the evaluation of aggregation propensity. We conducted MD simulation for 319 peptides and used them for training a machine-learning model to search all tetra-peptides. Some peptides with high predicted aggregation propensity were validated by MD simulation.

3Pos034 Molecular evolution of the structure elements in the TIM barrel family proteins

Yasumichi Takase[1], Yugo Hayashi[1], Yoichi Yamazaki[1], Sachiko Toma-Fukai[1], Hironari Kamikubo[1,2] ([1]*Div. Mat. Sci., NAIST*, [2]*IMSS KEK*)

Previously, we had identified essential segments for the tertiary structure of a protein from an amino acid sequence, termed structure elements, by comprehensive Ala-insertion mutation analysis. We hypothesized that structure elements worked as a kind of essential building blocks of the structure. This idea raises the question: are the elements conserved through evolutionary history? In this study, we focused on the TIM barrel superfamily and studied the conservation within some of the subfamilies. In the result, it was suggested that the positions and the length of the predicted elements were well conserved. From these results, it can be proposed that structure elements of the proteins are highly conserved and act as the building blocks of the TIM barrel motif.

3Pos035 メタノール中アラメチシンのヘリックス構造熱安定性
Thermal stability of helical conformation of alamethicin in methanol

Yoshinori Miura (*Center for Advanced Instrumental Analysis, Univ. Kyushu*)

Alamethicin is an ion channel peptide composed of 20 amino acid residues. Alamethicin has high solubility in methanol, while it is insoluble in water. In addition, the peptide has been known to form a helical conformation in methanol. We examined thermal stability of the helical conformation in the light of amide protons involved in intra-molecular hydrogen bonds by using proton NMR spectroscopy. We found that the helical conformation is maintained up to 54 C at least.

3Pos036 ヨウ素染色を用いた種の異なるインスリンアミロイド構造の識別及びシード依存的構造伝播の追跡
Structural difference and its seed-dependent propagation of human/bovine insulin amyloid fibrils as detected by iodine staining

Keisuke Yuzu, Eri Chatani (*Grad. Sch. Sci., Kobe Univ.*)

Amyloid fibrils are associated with many diseases and exhibit various structures showing different phenotypic properties. We previously discovered that iodine staining showed different colors depending on amyloid polymorphs. In this study, we investigated the iodine staining of insulin amyloid fibrils of different insulin species, human and bovine. As a result, they showed distinct absorption spectra shapes in visible region. Moreover, the spectral shape was conserved even after self-seeding and cross-seeding, suggesting that structural propagation occurred beyond fibril generations and furthermore, beyond species. This structural discrimination based on iodine staining can be a powerful tool for studying structural propagation mechanism of amyloid fibrils.

3Pos037 Examination of the possibility of the formation of transmissible transthyretin amyloid fibrils by the use of proteolysis

Misato Matsumura[1], Naoki Yamamoto[2], Keiichi Yamaguchi[3], Masatomo So[3], Yuji Goto[3], Eri Chatani[1] ([1]*Grad. Sch. Sci., Kobe Univ.*, [2]*Fac. Med., Jichi Med. Univ.*, [3]*Inst. Protein Res., Osaka Univ.*)

Transthyretin (TTR) is an amyloidogenic protein that causes senile amyloidosis. Studies on the mechanism of fibril formation of TTR have been conducted for many years, but it is difficult to obtain transmissible amyloid fibrils in vitro. In this study, we examined the propensity of wild-type TTR to form amyloid fibrils in the presence of trypsin or V8 protease. As a result of incubation in weak acidic conditions, slight increases in thioflavin T (ThT) fluorescence were observed with an optimal concentration of protease. The products obtained appeared to have seeding ability judging from the result of ThT fluorescence and circular dichroism spectra. It was suggested that certain characteristic truncations might be involved in amyloid fibril formation.

3Pos038 The mechanisms underlying the inhibition of amyloid formation by polyphenol

Yuto Kimura, Masatomo So, Yuji Goto (*IPR, Osaka Univ.*)

Many natural products including polyphenol have been reported to prevent amyloid formation, which is related to various diseases including neurodegenerative diseases. It is important to identify the mechanisms underlying the inhibition in order to prevent the diseases. In this study, we investigated the effects of kaempferol-7-O-glucoside (KG), which is one kind of polyphenol, on amyloid formation of α-synuclein (aSN) associated with Parkinson's disease. We showed that KG inhibited aSN fibril formation depending on its dose. Interestingly, the required amount of KG to prevent amyloid fibril decreased as the salt concentration increased. The results suggest that hydrophobic interaction between KG and protein has an important role in suppressing amyloid formation.

3Pos039 In vitro assembly of metabolon by liquid-liquid phase separation

Tomoto Ura, Kentaro Shiraki (*Pure and Appl.Sci.,Univ.Tsukuba*)

Metabolon is a multienzyme complex that is ubiquitous in metabolic pathways. Previous studies show that metabolon activates specific reaction by assembling specific enzymes needed. However, these studies have been only performed in vivo experiment; hence, the molecular mechanism of metabolon is elusive. Here, we hypothesize that liquid-liquid phase separation (LLPS) plays an important role for the formation of metabolon that regulates sequential enzyme reactions. In the presence of ATP condensed droplet by LLPS, sequential enzyme reaction that consume ATP is activated and sequential enzymes dynamically assemble in droplet. Our results suggest that metabolon is formed by specific enzymes and substrates condensed droplet.

3Pos040 Binding properties of heart-type Fatty-Acid-Binding Protein proved by 1,8-ANS displacement assay

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Fatty-Acid-Binding Proteins (FABPs) play essential roles in the cytoplasm to transport fatty acids (FAs) to the mitochondria for energy production and membrane proteins for signal transduction. In this study, binding properties of human heart-type FABP (FABP3) with C6-C16 saturated FAs were proved by 1,8-ANS displacement assay. Even after the addition of a sufficient amount of FAs, 1,8-ANS molecules remained in the binding pocket in an acyl chain length-dependent manner. The index of EC50 for the binding affinity, which can be estimated from FA-displacement-induced intensity changes in 1,8-ANS fluorescence, significantly decreased as the acyl chain of FAs becomes longer. It was also suggested that short-chain FAs coexist with 1,8-ANS in the FABP3 binding pocket.

3Pos041 混雑環境系におけるタンパク質ーリガンド結合機構の速度論的解析
Crowder effects on a protein-ligand binding process

Kento Kasahara[1], Suyong Re[1], Hiraku Oshima[1], Isseki Yu[4], Grzegorz Nawrocki[5], Michael Feig[5], Yuji Sugita[1,2,3] ([1]*RIKEN BDR*, [2]*RIKEN R-CCS*, [3]*RIKEN CPR*, [4]*Maebashi Inst. of Tech.*, [5]*Michigan State Univ.*)

Protein-ligand binding is one of the key processes in biological systems. In a cell, many non-target proteins and metabolites exist around the target protein and ligand molecules. The effect of non-target proteins is not well understood both experimentally and theoretically. In the present study, we investigate the non-target protein effects on the binding based on molecular dynamics simulation. Src kinase, PP1 inhibitor, and 8 BSA are used as a target protein, a ligand molecule, and non-target proteins. We found that minor structural changes in the kinase due to the interaction with BSAs change the binding pathways and mechanisms significantly. To examine the non-target protein effect, we carried out free-energy analysis after the sufficiently long MD simulations.

3Pos042 マニフォールドラーニングを用いた低温電子顕微鏡 4 次元イメージング法の確立に向けて
Toward constructing a four-dimensional imaging technique for cryo-electron microscopy with manifold learning

Takashi Yoshidome (*Department Applied Physics, Tohoku University*)

Elucidating the dynamics of proteins is a next generation of analyses of cryo-electron (cryo-EM) microscopy data. To achieve this, it is now desirable to construct a four-dimensional imaging technique. Here we present our recent simulation studies for a cryo-EM microscopy experiment toward a construction of a four-dimensional imaging technique based on the manifold learning. We will show that unlike previous classification techniques, the manifold learning enables us to find images between the states as well as those in the states. The finding of images between the states is inevitable toward the four-dimensional imaging technique.

3Pos043 投影方向に偏りがある低温電子顕微鏡像の全投影方向の像の回復に向けて
Toward a recovery of images from all irradiation directions using cryo-electron microscopy data with biased irradiation directions

Ryota Kojima, Takashi Yoshidome (*Department of Applied Physics, Tohoku University*)

Cryo-electron microscopy (Cryo-EM) experiment is currently one of the major structural analysis methods for proteins. An important assumption for the analysis is that projection images are obtained from a uniform direction. However, it is not always true, and in some case, we only have images from a biased direction. Here, we investigated whether the manifold learning can resolve the issue. We performed a simulation for a cryo-EM experiment, and obtained projection images from a uniform (data I) or a biased direction (data II). The manifolds of both data were projected onto a low-dimensional space. We found a condition that the manifold of the data II is equal to that of the data I, suggesting a possibility to resolve the issue.

3Pos044 バイオ医薬品のプロセス開発および QC における MAM のためのワークフロー駆動型プラットフォームソリューション
A workflow driven platform solution for MAM-based critical quality attribute monitoring of biotherapeutics in process development and QC

Kenji Hirose[1], Maki Terasaki[1], Shota Nakamura[1], Nilini Ranbaduge[2], Henry Shion[2], Ying Qing Yu[2], Min Du[2], Weibin Chen[2] ([1]*Nihon Waters*, [2]*Waters*)

Peptide-based Multi-Attribute Methods (MAM) that utilize Liquid Chromatography-High Resolution Mass Spectrometry (LC-HRMS) technology to simultaneously assess multiple potential Critical Quality Attribute (pCQAs) are being implemented in biopharmaceutical labs to reduce analytical complexity and increase productivity. Here we demonstrate the capabilities of a workflow-driven analytical platform comprised of a Time-of-Flight (TOF) MS for CQA identification, monitoring, and new peak detection. This LC-UV-TOF system has a compact laboratory footprint and provides integrated compliance-ready workflows for pCQA/CQA analysis and monitoring throughout the product life cycle.

3Pos045 カチオンイオン交換と超小型飛行時間型質量分析計を用いたオンライン IEX-MS による モノクローナル抗体チャージバリアントの特性解析とモニタリング
Online IEX-MS Characterization and Monitoring of mAb Charge Heterogeneity Us ing an Optimized Cation Exchange Resin and Compact TOF MS

Maki Terasaki[1], Shota Nakamura[1], Kenji Hirose[1], Samantha Ippoliti[2], Wang Qi[2], Yu Ying Qing[2], Lauber Matthew A.[2] ([1]*Nihon Waters K. K.*, [2]*Waters Corporation*)

Ion exchange chromatography (IEX) is a method of choice for the analysis of charge heterogeneity encountered with biotherapeutics drug candidates. In this study, MS-compatible ion exchange separations are combined with a new benchtop TOF MS instrument and applied to a case study on identifying the charge variants formed upon forced degradation. Using this direct IEX-MS method, separations of charge variants species were achieved at both intact and subunit-level of analysis. For the untreated antibodies, anticipated charge variants such as deamidation and C-terminal lysine variants have been successfully separated and identified. At subunit level, further separation and identification of additional N-glycoforms in the Fc region has also been achieved.

3Pos046 DFA イオンペア試薬、高密度フェニル結合充填剤による ADC の高感度 LC-MS プロファイリング
High sensitivity LC-MS profiling of ADC with difluoroacetic acid ion pairing and a high coverage phenyl-bonded stationary phase

Hiroko Iwasaki[1], Kenji Hirose[1], Jennifer Nguyen[2,3], Jacquelynn Smith[4], Olga V. Friese[4], Jason C. Rouse[4], Daniel P. Walsh[2], Matthew A. Lauber[2] ([1]*Nihon Waters K. K.*, [2]*Waters Corporation*, [3]*Univ. of Copenhagen*, [4]*Biotherapeutics Pharm, Sci., Pfizer WRD*)

A new method has been developed to enhance LC-MS subunit profiling of mAbs and ADCs that is based on the use of highly purified DFA as a mobile phase ion pairing agent and a high-coverage phenyl stationary phase. LC-MS quality DFA has been successfully prepared as verified through ICP metal quantitation and LC-MS application testing. DFA confers notable gains in MS sensitivity versus TFA, even at higher concentration, while providing comparable resolution. The method was proven to be robust and suitable for both UV and MS characterization of ADCs when qualified on three separate cysteine-conjugated ADCs.

3Pos047 ポリリン酸による鶏リゾチームの凝集
Polyphosphate-induced aggregation of hen lysozyme

Kenji Sasahara, Keiichi Yamaguchi, Masatomo So, Yuji Goto (*IPR Osaka university*)

Polyphosphate (PolyP) was recently reported to promote amyloid formation of several proteins. However, the mechanism underlying PolyP-induced amyloid formation is not well understood. Here, we examined PolyP-induced aggregation of hen egg-white lysozyme under acidic conditions. Our results show that near the solubility limit modulated by PolyP, the protein molecules in the labile state are highly prone to convert to amyloid fibrils. The calorimetric data suggested a crucial role of the dehydration of the protein molecules in the aggregation process. These results offer a model of protein aggregation modulated by the entropy-driven binding reaction of PolyP molecules, followed by the compensatory enthalpy-driven protein aggregation.

3Pos048 超小型高分解能質量分析計の導入によるバイオ医薬品開発の課題の解決
Meeting Challenges of Implementing Accurate-Mass Mass Spectrometry for Biotherapeutic Development in Regulated/non-Regulated Environments

Shota Nakamura[1], Taiji Kawase[1], Maki Terasaki[1], Kenji Hirose[1], Henry Shion[2], Mellisa Ly[2], Nilini Ranbaduge[2], Ximo Zhang[2], Yun Alelyunas[2], Jonathan Pugh[2], Robert Lewis[2], Jill Lord[2], Mark Halifax[2], Nick Tomczyk[2], Dale Cooper-Shepherd[2], Laetitia Denbigh[2], Ying Qing Yu[2], Jason Rouse[3], Weibin Chen[2] ([1]*Nihon Waters K.K.*, [2]*Waters Inc.*, [3]*Pfizer Inc.*)

Experienced mass spectrometry (MS) users have long been required to manage instrument operation, data processing and interpretation when high resolution (HR) accurate-mass MS is deployed in the biopharmaceutical industry. Here, we report the development of a novel high performance bench-top orthogonal acceleration time-of-flight (oa-TOF) LC-MS system with simplified and optimized standard operation modes to deliver automated, accurate, and reproducible mass measurements for proteins, peptides, and released glycans. In this study, our experiment results demonstrate that the limit of detection (LOD), mass accuracy and decovoluted mass profiles of intact mAbs and ADC molecules were comparable to other HRMS MS systems (QTOFs).

3Pos049 珪藻殻への有用タンパク質提示発現による機能性材料開発
Protein display on the silica frustules of a marine diatom

Natsuki Onishi, Kensuke Nakajima, Yoshinori Tsuji, Yusuke Matsuda (*Dept.Biosci., Grad.Sch.Sci.Tech., Kwansei Gakuin Univ.,*)

Diatoms have silica frustules with fine mesoscopic structures. Silaffin is the frustules formation factor secreted into the silica deposition vesicle (SDV). In this study, excretion of a small peptide to SDV were attempted. DNA sequence encoding a peptide, protamin tagged with GFP harboring SDV signal (TpSil3) and FLAG were constructed and expressed in diatom. The expressed TpSil3:FLAG:Protamine:EGFP was localized in the cytosol, while TpSil3:FLAG:EGFP:Protamine were localized in the frustule, indicating that highly basic amino acid cluster between Sil3 and GFP inhibited protein sorting into SDV. Interestingly, mutant with deleted arginine of C-terminus protamine was localized in the frustule, suggesting that C-terminal positive charge is responsible for mislocation.

3Pos050 Development of the engineered trimeric single-chain Fv fragment of the therapeutic antibody

Takashi Tadokoro, Kota Nakamura, Harumi Tsuboi, Katsumi Maenaka (*Faculty of Pharmaceutical Sciences, Hokkaido University*)

Antibody drugs are attractive as effective therapeutic agents. However, antibody can sometimes bind to normal tissue which slightly express the target molecule with its high specificity and affinity, that leads side effects. Here, we designed and prepared the novel antibody fragments, which consist of an engineered trimeric single-chain Fv antibody fragment (scFv). It is expected that high specificity and affinity to the target can be achieved by the avidity effect. For this, affinity suppressing mutations were introduced into the scFv of the therapeutic antibody, and the scFv mutants were multimerized by addition of the coiled-coil domain at the C-termini of the scFv. We will present and discuss about the physicochemical properties of the trimeric antibody fragments.

3Pos051 多様なユビキチン鎖のロバストな合成法
Robust synthesis methods of various ubiquitin chains

Takumi Suzuki, Takahiro Aizu, Yutaka Ito, Masaki Mishima (*Grad. Sch. Sci., TMU*)

It is known that the intracellular role of polyubiquitination differs depending on the polyubiquitination site. In addition, it has been found that polyubiquitin chains link in not only a homogenous binding mode but also mixed linkage and branched binding mode. Branched polyubiquitin chains have only partially understood, and thus more detailed analyses are required. To facilitate these studies, we offer synthesis methods for branched polyubiquitin chains based on chemical biology without E1 to E3 enzymes. We used a click reaction and a protein ligation reaction by sortase A. and succeed in synthesizing branched tetrameric ubiquitin. In this presentation, we will report the actual synthetic strategy, and the improved synthetic strategy.

3Pos052 De novo design of protein structures with Ploop-motif for ATP binding

Hiroko Yamada[1], Kengo Nakamura[1], Takahiro Kosugi[1,2,3], Nobuyasu Koga[1,2,3] ([1]*SOKENDAI*, [2]*NINS IMS*, [3]*NINS ExCELLS*)

One of the goals for protein design is to create functional proteins from scratch. Recently, we discovered general principles for designing protein structures, which enabled de novo design of various protein structures without function. Here, for designing functional proteins from scratch, we attempt to create protein structures with a binding motif for a small molecule. Especially, we aimed to design protein structures with P-loop motif for ATP.

3Pos053 金属タンパク質における酸化還元電位の第一原理計算法の開発
An ab initio method of evaluating redox potential for metalloprotein

Cheng Cheng, Shigehiko Hayashi (*Kyoto Univ*)

Redox processes are involved in various electro-chemical systems as fuel cell, photosynthesis, energy storage, and enzymes. Redox reactions which occur in complicated bio-systems consisting of flexible proteins, mobile solvent water molecules, and lipids are coupled to their thermal fluctuations, and provide electro-chemical properties different from that in water solutions. In this study, we evaluated the redox potential of cytochrome c by calculating free energy difference between the redox states. The calculated redox potential is 107.4 kcal/mol ,which is overestimated only by 0.6 kcal/mol compared to the experimental value of 108.0 kcal/mol. This ab initio approach thus allows one to readily evaluate redox potentials of metalloprotein systems.

3Pos054 ウシミトコンドリア由来酸素還元酵素の活性型の単量体構造
Monomeric structure of an active form of respiratory oxygen reductase from bovine mitochondria

Kyoko Shinzawa-Itoh[1], Takashi Sugimura[2], Tomonori Misaki[2], Yoshiki Tadehara[1], Shogo Yamamoto[1], Makoto Hanada[1], Naomine Yano[1], Tetsuya Nakagawa[3], Shigefumi Uene[1], Takara Yamada[3], Hiroshi Aoyama[4], Eiki Yamashita[5], Tomitake Tsukihara[1,5], Shinya Yoshikawa[1], **Kazumasa Muramoto**[1] ([1]*Graduate School of Life Science, University of Hyogo*, [2]*Graduate School of Material Science, University of Hyogo*, [3]*School of Life Science, University of Hyogo*, [4]*Graduate School of Pharmaceutical Sciences, Osaka University*, [5]*Institute for Protein Research, Osaka University*)

Respiratory chain generates proton motive force coupled to electron transfer at high energy efficiency. Respiratory O_2 reductases are broadly classified into A, B and C-types based on their molecular structures. A-type O_2 reductase isolated from bovine mitochondrial membrane exists as a dimer in the previous crystal structures, whereas it exists as a monomer in the respiratory supercomplex. Other O_2 reductases exist as a monomer. In this study, we prepared monomeric and dimeric forms of bovine O_2 reductase, stabilized using amphipol, and showed that the monomer had high activity. Using a newly synthesized detergent, we determined the oxidized and reduced structures of monomer with resolutions of 1.85 Å and 1.95 Å, respectively.

3Pos055 脂質・コレステロール・タンパク間の協同性による上皮成長因子受容体の膜近傍ドメイン２量体形成機構
Lipid-cholesterol-protein interaction in the dimerization of juxtamembrane domains of epidermal growth factor receptor

Ryo Maeda[1], Yasushi Sako[1], Takeshi Sato[2] ([1]*Cellular Informatics Lab., RIKEN*, [2]*Kyoto Pharmaceutical Univ.*)

Transmembrane helix and juxtamembrane (JM) domains bridge the extracellular and intracellular domains of epidermal growth factor receptor, EGFR. Although JM dimerization is thought to play a crucial role in regulation of EGFR kinase activity, the underlying kinetic mechanisms remain unclear. Here, combining single-pair FRET imaging and nanodisc techniques, we showed that electrostatic interactions between basic residues and negatively charged lipids facilitated JM dimerization effectively. Furthermore, we found that cholesterols in the membrane increased FRET efficiency between JM protomers and induced transitions from monomer to dimer conformations. These results indicated that cholesterols can help the assembly of JM domains more closely, related to EGFR clustering.

3Pos056 バクテリオルベリン及びその前駆体の膜タンパク質ハロロドプシンへの結合特異性
Binding specificity of bacterioruberin and its precursors to membrane protein halorhodopsin

Fumiya Hattori, Takanori Sasaki (*Grad.Sch.Adv.Math.Sci.,Meiji Univ*)

A light driven anion pump halorhodopsin from N.pharaonis (NpHR) forms trimer on the membrane, and specifically binds a carotenoid of bacterioruberin(BR) at the crevice between the protein molecules. So far, the molecular recognition mechanism for NpHR-BR complex formation has been unclear. In this study, NpHR obtained from E.coli over-expression system and carotenoid components extracted from archaeal membrane were mixed in detergent solution, and the carotenoids involved in complex formation were analyzed by HPLC. As a result, BR and four kinds of BR precursors has formed complex with trimer NpHR. This result suggests that NpHR has the ability to recognize and bind even the BR precursors with lower number of OH groups at the end groups than that of BR.

3Pos057 細胞膜上 Akt の１分子イメージングによるシグナル伝達機構の研究
A single molecule imaging approach to understand signal transduction mechanism through Akt on the plasma membrane

Hideaki Yoshimura, Takeaki Ozawa (*Sch. Sci., Univ. Tokyo*)

We studied on signal transduction mechanisms of Akt, a signaling “node” molecule. Akt initially localizes in the cytosol, and translocates onto the plasma membrane upon PIP3 production in the the membrane after extracellular stimulations. To understand the mechanism of the signal transduction through the node molecule Akt, We prepared Akt-EGFP and introduce it into HeLa cells. The single molecule dynamics of Akt-EGFP was analyzed and the number of molecules and residency times on the plasma membrane were estimated. Based on the results, we will discuss the mechanism of signal transduction through Akt especially of the specificity in phosphorylation of downstream molecule depending input signals.

3Pos058 Triton X-100 により可溶化した Proteorhodopsin の光機能中間体の速度論的解析
Kinetic analysis of photointermediates of Proteorhodopsin solubilized with Triton X-100

Airi Yamamoto[1], Fumio Hayashi[2], Takashi Kikukawa[3,4], Masashi Sonoyama[1,5,6] ([1]*Grad. Sch. Sci. Tech., Gunma Univ.*, [2]*Inst. Anal. Cent., Gunma Univ.*, [3]*Fac. Adv. Life Sci., Hokkaido Univ.*, [4]*GI-CoRE, Hokkaido Univ.*, [5]*GIAR., Gunma Univ.*, [6]*GUCFW., Gunma Univ.*)

Proteorhodopsin (PR), is a light-driven proton pump similar to a well-studied membrane protein bacteriorhodopsin. In this study, Triton X-100 (TX-100) was employed for solubilization of PR instead of a popular detergent, n-dodecyl-β-D-maltoside (DDM). The incubation of PR with TX-100 at the concentration of 1.0% for ~50 h resulted in successful solubilization at the level comparable to the solubilization with DDM. Comparative measurements on solubilized PR with TX-100 and DDM by using laser flash photolysis demonstrated that the photocycle of the TX-100-solubilized PR is longer than that of the DDM-treated PR. Kinetic properties of photointermediates, which are under investigation by global fitting analysis, will be reported.

3Pos059 1 分子イメージングによる TRPV1 チャネル・脂質間相互作用の時空間動態解析
Spatiotemporal analysis of TRPV1 channel-lipid interaction by single molecule imaging

Yutaro Kuwashima[1,2], Masataka Yanagawa[2], Mitsuhiro Abe[2], Yasushi Sako[2], Ryohei Aoyagi[1], Makoto Arita[1] ([1]*Grad. Sch. Pharm., Univ. Keio*, [2]*Wako Inst., Riken*)

TRPV1 is a nociceptor channel that is activated by various stimuli, such as capsaicin, heat, and inflammatory mediators. The channel activity of TRPV1 is also regulated by membrane lipids including PIP2. However, the spatiotemporal organization of TRPV1 and lipids in living cells is poorly understood. Here we show that immobile TRPV1 molecules observed by single-molecule imaging increased after capsaicin stimulation at lower concentrations than those induce Ca2+ influx and endocytosis of TRPV1. Dual-color colocalization analysis suggested that the affinity of TRPV1 for PIP2 decreased upon capsaicin binding. Because PIP2 is a positive regulator of TRPV1, the immobilization of TRPV1 and spatial segregation from PIP2 would play a key role in the desensitization of TRPV1.

3Pos060 The effect of the distance between the RNA sequences recognized by two RNA-binding domains on the affinity of the MSI1-RNA interaction

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Musashi-1 (MSI1) plays key roles in self-renewal of stem cells and also involved in tumorigenesis by regulating the translation of target mRNAs. MSI1 reportedly binds to the 3'-untranslated regions (3'UTRs) of various target mRNAs. MSI1 contains two RNA binding domains (RBDs), RBD1 and RBD2. Here, we investigate how the distance between the two sequences each recognized by RBD1 and RBD2 would affect the target RNA recognition by MSI1. We evaluated the binding affinities of MSI1 against various RNA sequences derived from numb 3'UTR, a well-established MSI1 target, by FA and EMSA. MSI1 turned out to bind to RNAs of different lengths with distinct affinities. How MSI1 recognizes RNA of different lengths will be discussed.

3Pos061 Computational insights into DNA binding affinity and its repair activity for photolyase/cryptochrome superfamily

Ryuma Sato[1], Yoshiharu Mori[2], Noriaki Okimoto[1], Makoto Taiji[1] ([1]*RIKEN*, [2]*Kitasato univ*)

Anacystis nidulans cyclobutane pyrimidine dimer photolyase (CPD-PL) and Arabidopsis thaliana cryptochrome-DASH (CRY-DASH) are blue-light photoreceptors which involve the flavin adenine dinucleotide. CPD-PL exerts function which repairs UV-damaged DNA, while CRY-DASH does not exert that, although CRY-DASH is high sequence homology with photolyases. To elucidate the reason why CRY-DASH cannot repair UV-damaged DNA, we investigated a behavior to DNA of CPD-PL and CRY-DASH in terms of the DNA binding using molecular simulations. We found the important difference between CPD-PL and CRY-DASH for DNA binding affinity, and this result might be related to DNA repair activity. We will explain this with relevance to DNA repair activity in our presentation.

3Pos062 The directionality regulation mechanism of serine recombinase

Hsiu-Fang Fan (*NSYSU*)

Serine integrases promote recombination between two different DNA sites and its directionality is regulated by the recombination directionality factor (RDF). Here, we utilize tethered particle motion (TPM) experiment to investigate the regulation role of coiled-coil domain in PhiC31 integrase-mediated site-specific recombination. Fast escapes from the non-functional side pathway and preferentially correct alignment in synapse state were observed for 449K mutants, leading to hyperactive recombination efficiency. Moreover, two Int E449K dimers are required for attL x attR recombination in the absence of RDF. FCS data reveals the short lifetime of the productive synaptic complex, supporting the gate-rotation model.

3Pos063 Single-molecule studies of how polyamines stimulate RecA-mediated recombination

Naciye Esma Tirtom[1], Yang Hsu[2], Hung-Wen Li[1] ([1]*NTU*, [2]*NTNU*)

Polyamines are naturally occurring cationic molecules and were reported to stimulate RecA-mediated DNA recombination, but the molecular mechanism is not characterized yet. We utilized single-molecule tethered particle motion (TPM) experiments to investigate this phenomenon. We first determined the condensation efficiencies for 2 natural polyamines, spermine and spermidine. Later, using invading strand experiments, we showed addition of polyamines resulted in a longer-dwell time intermediate and higher number of transient tethers, which together increased the D-loop formation by 18 times. Via electrostatic interactions with DNA, polyamines sufficiently increase the contact between RecA nucleoprotein filaments and DNA, leading to stimulation of recombination progression.

3Pos064 (3SEA-3) 温度上昇とテラヘルツ光照射は転写反応に異なる影響を及ぼす。
(3SEA-3) Terahertz radiation and temperature increase differently affect transcription by RNA polymerase

Masahiko Imashimizu[1], Masahito Tanaka[1], Hiromichi Hoshina[2], Koh Takeuchi[1] ([1]*AIST*, [2]*RIKEN*)

Thermally driven fluctuations play a critical role in key cellular processes such as DNA replication and transcription. Recent spectroscopic studies have shown that such fluctuating motions of protein and DNA include collective vibrational modes, which are detected as sharp peaks in the terahertz (THz) frequency region. In this study, the effects of THz radiation on transcription by RNA polymerase were analyzed by high-throughput sequencing. We demonstrated that THz radiation enhanced transcription pausing (~kT process), while temperature rise without THz radiation accelerated escape of the paused complex to elongation (>>kT process). This result suggests that the THz radiation specifically affected the biochemical reaction in a manner separable from thermal effects.

3Pos065 液体状 DNA の相分離を利用したドメインを持つマイクロゲルカプセルの構築
Construction of hydrogel microcapsules with domain by using phase separation of liquid-like DNA

Yuji Nakashima, Yusuke Sato, Masahiro Takinoue (*Dept. of Comp. Sci., Tokyo Tech.*)

Microcapsules that can encapsulate materials inside are used for biomedical applications, molecular robots, and so on. However, functions that do not only keep encapsulated materials of the microcapsules such as recognition and response to an external stimulus are still developing. DNA hydrogel is a good technique to construct micro-sized structures and has various stimulus responses; therefore, we apply this technique for construction of microcapsules. In this study, we construct DNA hydrogel microcapsules with domain at a water-in-oil droplet interface using two kinds of Y-shaped DNA nanostructures designed to phase separate. These microcapsules will have multiple functions on domain and be applied in various fields.

3Pos066 DNA 液滴を用いた液-液相分離による動的システム
Dynamic system by liquid-liquid phase separation using DNA droplet

Nozomi Tsumura, Yusuke Sato, Yuji Nakashima, Masahiro Takinoue (*Tokyo Tech*)

Inside the cell nucleus, there are two states of DNA: expanded euchromatin structure with high transcriptional activity and condensed heterochromatin structure without transcription. These states are expected to be formed by liquid phase separation. Phase separation is a phenomenon in which a mixture of two or more substances separates into each component. By imitating such a dynamic system using DNA gel, we consider that it becomes possible to elucidate the system of cell nucleus and to design an artificial molecular machine. We aim to reproduce a dynamic system by creating a DNA droplet that generates phase separation inside the DNA droplet by an external stimulus.

3Pos067 DNA 液滴ポーラスマイクロ構造の形成
Porous microstructure formation of DNA droplet

Tetsuro Sakamoto, Yusuke Sato, Masahiro Takinoue (*Tokyo Institute of Technology*)

Self-organized microstructure of programmed DNA molecules is one of the best materials for making microrobots or artificial cells. We are particularly studying porous DNA-microstructure for microrobots body or artificial cell nucleus. This kinds of complex structure and self-organizing process might be also important to understand inter-cellular life system. In this study, we focused on a phenomenon of microporous formation of DNA solution and we discovered a simple and stable process of porous DNA-microstructure formation. This material and process could be valuable for not only material use of microrobots or artificial cell but also understanding complex life system.

3Pos068 DNA ゲルの相転移のシミュレーションモデルと解析
Simulation model and analysis of phase transition of DNA gel

Akihiro Yamamoto, Tetsuro Sakamoto, Yusuke Sato, Masahiro Takinoue (*School of Computing, Tokyo Institute of Technology*)

In living cells, phase separation of biomolecules is expected to play important roles. Understanding such the phase separation is important not only for biology but also for molecular robotics, which aims to nano/microrobots inspired by living systems. In this study, we use DNA hydrogels, which have three states: dissolved, droplet, and gel states. Here, we constructed a coarse-grained model and reproduce this the phase separation phenomenon of DNA gels by molecular dynamics simulation based on the model. We believe that this study will lead to the understanding of the mechanism of phase separation in living cells and then it will be applied to the development of new devices for molecular robot.

3Pos069 捩じれストレス下におけるヌクレオソーム DNA 解離および H2A/H2B2 量体脱離の自由エネルギープロファイル
Free energy profiles of unwrapping nucleosomal DNA under torsional stress and eviction of the H2A/H2B dimer

Hisashi Ishida, Hidetoshi Kono (*Institute for Quantum Life Science, QST*)

The structure and dynamics of chromatin is exposed to and affected by torsional stress in a nucleus. To understand how the torsional stress changes the stability of nucleosomes, we carried out all-atom molecular dynamics simulations to examine the unwrapping of the outer superhelical turn of the nucleosomal DNA from the histone octamer under the torsional stress imposed on the edges of the DNA. We found that the torsional stress can control the transitional path of symmetric and asymmetric unwrapping of the DNA by changing the interaction between the DNA and the histone core through conformational change in the DNA. We also show the free energy profile of eviction of the H2A/H2B dimer and conformational change of the histone core during the DNA unwrapping.

3Pos070 転写開始複合体における DNA 開裂に関連した DNA・タンパク質間相互作用の検討
DNA-Protein Interaction Related to DNA Opening in Transcription Initiation Complex

Genki Shino, Masahiro Shimizu, Shintaroh Kubo, Toru Niina, Shoji Takada (*Dept. of Biophys., Div. of Bio. Sci., Grad. Sch. of Sci., Univ. of Kyoto*)

The molecular mechanism of transcription initiation process has been actively studied by using cryo-electron microscopy and biochemical experiments. The transcription of eukaryotic protein-coding genes results from forming transcription initiation complex on the promoter DNA, consisting of RNA polymerase II and the six general transcription factors, and DNA opening. However, the details of DNA opening is unclear. To address this problem, we performed coarse-grained molecular dynamics simulations, and investigated how DNA in transcription initiation complex opens. We found that certain DNA-protein interactions result in the mechanism of DNA opening.

3Pos071 Poly(*N*-isopropylacrylamide)のコイル-グロビュール転移に伴う水への溶解度の劇的低下の物理
Physics of drastic decrease in water solubility upon coil-to-globule transition of poly(*N*-isopropylacrylamide)

Masao Inoue, Tomohiko Hayashi, Simon Hikiri, Masahiro Kinoshita (*Inst. Adv. Energ., Kyoto Univ.*)

Poly(*N*-isopropylacrylamide) (PNIPAM) is in water-soluble coil state below a critical temperature T_C and in water-insoluble globule state above T_C. We consider structural ensembles of the coil and globule states generated below and above T_C, respectively. Using our recently developed method for calculating the hydration free energy (HFE) of a polyatomic solute, we show that the HFEs of the coil and globule states take large negative and positive values, respectively. Decomposition of the HFE into a variety of energetic and entropic components provides us with physical insights into the remarkable difference between the two states in water solubility. Thermodynamic properties of PNIPAM in the two states are compared with those of a protein in unfolded and folded states.

3Pos072 Water-protein interactions coupled with protein conformational transition

Soichiro Kitazawa[1], Takuro Wakamoto[2], Ryo Kitahara[1] ([1]*College of Pharmaceutical Sciences, Rtisumeikan Univ.*, [2]*Graduate School of Life Sciences, Ritsumeikan Univ.*)

Water-protein interactions can be altered by conformational transition of a protein. We investigated pressure-induced changes in exchange rates of water-to-amide proton by two NMR approaches, high-pressure phase-modulated clean chemical exchange and high-pressure H/D exchange. Water-amide proton exchange rates of residues 32-35, 40, 41 and 71 of ubiquitin were significantly increased with the transition from its native state (N1) to an alternative state (N2). These results indicate the destabilization of the hydrogen bonds in the backbone and partial exposure of those amide groups to solvent in N2. Global unfolding was detected by high-pressure H/D exchange. The current method is useful to describe the entire energy-landscape of a protein.

3Pos073 SASA モデルにおける蛋白質間相互作用の過安定化
Over-stabilization of protein-protein interaction in solvent accessible surface area model

Kohei Kuroishi, Dan Parkin, Akira Yodogawa, Mitsunori Takano (*Dept. of Pure & Appl. Phys., Waseda Univ.*)

The solvent-accessible surface area (SASA) model, in combination with the generalized Born (GB) model, is widely used in molecular dynamics simulations. In the GB/SASA model, the nonpolar term of solvation free energy is proportional to SASA with a positive coefficient γ, favoring the bound state. Previously, we found that barnase-barstar binding was over-stabilized in the GB/SASA model presumably due to the nonpolar term of solvation free energy. In this study, we investigated the effect of γ on the barnase-barstar binding. We found that a binding-unfavorable negative γ is suitable, in contrast to the widely used positive γ derived from alkane experiments. We will present the physical reason for the negative γ.

3Pos074 Machine-learning approach for water dynamics

Taku Mizukami[1], Viet Cuong Nguyen[3], Hieu Chi Dam[2] ([1]*JAIST, Materials*, [2]*JAIST, Knowledge*, [3]*HPC.Inc*)

Water is a ubiquitous material and plays an important role in many fields. Biomolecules are functionally activated under a hydrated environment, by mechanisms that are influenced by interactions with hydration water. In this study, we demonstrate a machine-learning approach to predict the physicochemical features of water dynamics. The chemical environment of atom was defined and estimated by coordinates calculated by molecular dynamics simulations of bulk water system and/or solute-solvent system. The sparse modeling techniques, like lasso or lars, were applied to leaning and prediction on the physicochemical features of water. We will discuss the prediction for local environment of water cluster.

3Pos075 (3SFA-7) 三次元構造モデルから発生過程における細胞機能の理解を試みる
(3SFA-7) Attempt to understand the cellular function during developmental process from 3D structural model

Junpei Kuroda[1,4], Takeshi Itabashi[1,2,3], Takako M. Ichinose[1], Shigeru Kondo[4], **Atsuko H. Iwane**[1,2,3] ([1]*Cell Field Struc., BDR, Riken*, [2]*Grad. sch. Integ. Sci. Life, Hiroshima Univ.*, [3]*Spec. Res. Promot. Group, Grad. Sch. Fronti., Biosci., Osaka Univ.*, [4]*Pattern formation, Grad. Sch. Fronti., Biosci., Osaka Univ.*)

In order to understand how tissue complex morphology is formed in developmental process, it is necessary to reveal three-dimensionally the positional information and morphology information of individual cells correlated of cell biological analysis. Therefore, we decided to try a method using FIB-SEM and 3D-reconstruction techniques to understand the morphogenesis mechanisms of tissue and the cellular functions in developmental process. We focused on collagen crystals called actinotrichia, which are thought to play a central role in zebrafish fin formation. In this meeting, we will present the interaction between actinotrichia and the cells surrounding it involved in a three-dimensional manner during the fin development, and discuss the cell functions from our 3D models.

3Pos076 脳形態形成におけるニューロン移動と大脳成長の連成数理モデリング
A coupled mathematical modeling for neuronal migration and cerebral growth in brain morphogenesis

Hironori Takeda[1], Yoshitaka Kameo[1,2,3], Taiji Adachi[1,2,3] ([1]*Grad. Sch. Eng, Kyoto Univ.*, [2]*Inst. Front. Life Med Sci., Kyoto Univ.*, [3]*Grad. Sch. Biostudies, Kyoto Univ*)

During cerebral morphogenesis, neurons born in a ventricular zone migrate towards their proper position to form a layered structure. In this process, a cerebral tissue deforms due to growth driven by mechanical effects of neuronal activities. In this study, in order to investigate the interaction between neuronal activities and tissue growth, we developed a coupled model for neuronal migration and tissue growth based on continuum mechanics. We performed finite element analysis to reproduce formation of layered structure in a growing cerebral cortex. This model will be helpful to understand how tissue morphologies and functional structure are formed depending on cellular activities.

3Pos077 Modeling of sea urchin gastrulation based on cytoskeleton imaging

Kaichi Watanabe, Naoaki Sakamoto, Akinori Awazu (*Integrated Sciences for Life in Hiroshima University*)

Gastrulation is one of the most important and universal processes for morphogenesis. Sea urchin embryo is the typical model organism of morphogenesis showing clear gastrulation process. We found H+/K+-ATPase ion pump activity play important roles to proceed the gastrulation; gastrulation was inhibited and embryo exhibited anomalous shape when the activity of H+/K+-ATPase was suppressed. In this study, we first construct a mathematical model of sea urchin embryo that can reproduce the normal and H+/K+-ATPase inhibition induced anomalous gastrulation processes. Based on the above model, and fluorescence imaging data of cytoskeleton, extracellular matrix, and pH, we investigate the inter- and intrarcellular chemo-mechanical coupling contributions to gastrulation.

3Pos078 細胞分裂に関わるキネシン 5 の頭部間協調におけるネック領域の役割の高速一分子観察
High-speed single molecule studies for the role of the neck region on the head-head coordination of mitotic kinesin-5

Taiga Yamada[1], Kohei Matsuzaki[2], Michio Tomishige[2] ([1]*Grad. Sch. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*)

Kinesin-5 is a motor protein that is involved in mitotic spindle formation. We previously reported that kinesin-5 moves processively under high ionic conditions, but the two heads are less coordinated compared to that of kinesin-1. To elucidate the underlying mechanism, we engineered a chimeric protein in which the entire stalk region was replaced with that of kinesin-1 and observed the motion using a high-speed dark field microscopy. We found that the chimeric motor showed improved head-head coordination (i.e., decreased rebinding rate), and the detached head remained unbound while waiting for ATP-binding, which is similar to that of kinesin-1. These results suggest that the neck coiled-coil is one of the reasons for the impaired head-head coordination of kinesin-5.

3Pos079 DNA ナノスプリングによる負荷を受けながら運動するキネシン 1 の高速一分子観察
High-speed single-molecule observations of kinesin-1 moving under a load from DNA origami nanospring

Kohei Matsuzaki[1], Mitsuhiro Iwaki[2], Michio Tomishige[1] ([1]*Dept. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*, [2]*BDR, Riken*)

Kinesin-1 is a motor protein that moves processively along microtubules. Stepping motion of kinesin-1 under load have been studied using optical trapping assays, although intermediate states during the motion have not been directly observed because of the technical difficulties. In this study, we observed unbinding/binding motions of a kinesin head under load using DNA origami nanospring with a high-speed dark-filed microscopy. We found that under substantial load, the unbound head diffuses but its position was moved backward. We also showed that the dwell time of the unbound state increased, and the frequency of the unbound head rebinding was increased. These results suggest that stepping rate is decreased because the head-head coordination is impaired under load.

3Pos080 Kinetic parameters and reaction scheme of high and low activity mutants of Serratia marcescens chitinase A

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Catalytic activity of Serratia marcescens chitinase A, a linear molecular motor, increases by two tryptophan mutations in the catalytic cleft (F232W/F396W) and decreases by single alanine mutation at the chitin binding domain (W69A). In this study, we investigated the properties of these mutants in detail by using biochemical and single-molecule analyses. Interestingly, these mutants showed comparable turnover rate (kcat) higher than WT. The high kcat of F232W/F396W related to its higher processivity. On the other hands, high dissociation rate after productive binding seems to be the reason of high kcat of W69A. We will propose and discuss a model which explains properties of these mutants and in which all experimentally determined kinetic parameters are included.

3Pos081 高速原子間力顕微鏡により観察された微小管上の外腕ダイニン
High-speed atomic force microscopy on outer dynein arms aligned on microtubules

Kenta Ishibashi[1,2], Kazuhiro Oiwa[2,3,4] ([1]*Osaka Univ.*, [2]*NICT-CiNet*, [3]*Advanced ICT Research Institute*, [4]*University of Hyogo*)

In Chlamydomonas axonemes, precisely-aligned dynein arms on the outer-doublet microtubules (MT) function in a coordinated fashion and produce periodic flagellar beating. To understand the coordination mechanism, we have investigated the dynamics of outer dynein arms (ODA) on a MT using a high-speed atomic force microscope (HS-AFM). We employed reconstituted MT-ODA complexes as well as frayed axonemes and equipped the commercially available HS-AFM with the fluorescence imaging system improved the efficiency of finding samples non-uniformly absorbed on the mica surface. The structural repeat of ODA was confirmed in the topology of MT-ODA complexes. The HS-AFM images thus defined detailed positions of axonemal components and will provide the dynamics of these components.

3Pos082 Development of novel Photochromic inhibitors for kinesin Eg5 which form multiple isomerization states utilizing azobenzene and spiropyran

Islam Md Alrazi, Kei Sadakane, Shinsaku Maruta (*Department of Bioinformatics, Graduate School of Engineering, Soka University, Hachioji, Tokyo , Japan*)

Mitotic kinesin Eg5 is an ATP driven motor protein. It is essential for the formation of bipolar spindles during eukaryotic cell division and is one of the most attractive targets for cancer treatment. In this study, to control mitotic kinesin Eg5, novel photochromic inhibitors have been synthesized which composed of photo-responsive azobenzene and Spiropyran derivatives. The significance of these multiple isomerization states is for clinical use and can control the therapy of the anticancer drugs more precisely. This compound showed multiple isomerization states upon the Visible (VIS) light, UV light and in the dark respectively. The synthesized compounds showed the different inhibitory activity of ATPase among their three different isomerization states.

3Pos083 回転子変異体を用いたべん毛モータースイッチ機構の解析
Analysis of the bacterial flagellar switch using mutant rotor components

Mai Kato[1], Tsubasa Ishida[1], Myu Yoshida[2], Yoshisyuki Sowa[1,2,3] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ.*,, [2]*Dept. Frontier Biosci., Hosei Univ*, [3]*Micro-nano Tech., Hosei Univ*)

Swimming of bacteria is powered by rotating their flagella. Their rotation is driven by a reversible rotary nano-machine, a bacterial flagellar motor, embedded in the cell membrane. Binding of a chemotactic protein CheY-P to rotor components induces motor switching from counterclockwise (CCW) and clockwise (CW) state. Here, we examined the characteristics of motor switching by mixing CCW- and CW-biased rotor proteins, FliG or FliM. Switching behaviors were monitored by a tethered cell assay or a bead assay. We also monitored the number of CCW- or CW-biased components incorporated into rotor rings by fusing fluorescent proteins. The correlation between the motor switching and ring assembly of CCW- and CW- components at single motor level will be presented.

3Pos084 細菌べん毛モーターの回転を支える回転軸-軸受間相互作用の解析
Analysis of shaft-bearing interactions that support the smooth rotation of bacterial flagellar motors

Yumi Kumazaki[1], Tsubasa Ishida[1], Myu Yosida[2], Yoshiyuki Sowa[1,2,3] ([1]*Grad. Sch. Sci. & Eng, Hosei Univ.*, [2]*Dept. Frontier Biosci., Hosei Univ.*, [3]*Micro-nano Tech., Hosei Univ.*)

The flagellar motor is embedded in the cell membrane, and spans the cell wall. Rod works as a shaft and connects a motor and an external structure of a flagellum. LP ring embedded in the cell wall supports smooth rotation of a rod as a bearing. The rod surface is negatively charged, and it was speculated that the electrostatic repulsion force between a rod and inner surface of LP ring may be important for its smooth rotation (Fujii *et al*. 2017). In this study, we investigated the interaction between a rod and LP ring by mutagenesis analysis and photo-crosslinking method. We found some negatively charged residues are crucial for the motor function. We also succeeded in detecting cross-linked products by substituting these residues to photo-reactive amino acid analogues.

3Pos085 Controlling the rotation speed of the bacterial flagellar motor with light-driven rhodopsin

So Hasegawa[1], Rei Abe-Yoshizumi[2], Keiichi Inoue[3], Hideki Kandori[2], Yoshiyuki Sowa[4] ([1]*Grad. Sch. Sci. & Eng., Hosei Univ*, [2]*Grad. Sch. Eng., Nagoya Inst. Tech*, [3]*Inst. Solid State Phys., Univ. Tokyo*, [4]*Dept. Frontier Biosci., Hosei Univ*)

The bacterial flagellar motor is a rotary nano-machine which converts the energy gained from the flux coupling ions across the cytoplasmic membrane to its rotation. Previous studies showed that a light-driven H^+ pump, proteorhodopsin, can be used to optogenetically control the speed of H^+-driven bacterial flagellar motor in *Escherichia coli* (Walter *et al*. 2007). In this study, we used a light-driven Na^+ pump, NdR2, to control sodium-motive force of *E. coli*. We observed the abrupt increase of speed of chimeric Na^+-driven bacterial flagellar motor in *E. coli* expressing NdR2 with light irradiation. The speed increases with light under various external sodium concentrations are under investigation.

3Pos086 *Paenibacillus* sp. TCA20 と大腸菌に由来するべん毛モーターキメラ固定子のイオン選択性
Ion specificity of chimeric stator proteins between *Paenibacillus* sp. TCA20 MotB1 and *Escherichia coli* MotB

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Bacterial flagellar motor consists of a rotor and multiple stator complexes. The transmembrane region (TM) of each stator complex functions as an ion channel and converts the flux of coupling ions to motor rotation. Stator complex MotA1MotB1 of *Paenibacillus* sp. TCA20 (TCA-MotAB1) was reported to use divalent cations as coupling ions. Here we engineered a chimeric stator protein between TCA-MotB1 and *E.coli* MotB. *E. coli* Δ*motAB* cells expressing TCA-MotA1 and the chimeric protein showed significant motility in the absence of divalent cations and monovalent cations excluding proton. The TM of the chimeric stator is derived from TCA-MotAB1. Therefore, the ion specificity of TCA-MotAB1 may be switched to be optimized for the membrane or external environments.

3Pos087 DNA オリガミを用いた野生型と変異体キネシン二分子による協調運動の観察
Cooperative transport by wild-type and mutant kinesin motors as studied by using programmable DNA origami

Shu Takano[1], Ryosuke Masuda[3], Kohei Matsuzaki[2], Mitsuhiro Iwaki[4], Michio Tomishige[2] ([1]*Grad. Sch. Sci. Eng., Aoyama Gakuin Univ.*, [2]*Dept. Math. Phys., Col. Sci. Eng., Aoyama Gakuin Univ.*, [3]*Dept. Appl. Phys., Grad. Sch. Eng., Univ. Tokyo*, [4]*BDR, Riken*)

Kinesin moves along microtubules to transport cargos inside the cells. Multiple motors coordinate to transport the cargo, but the underlying mechanism is still unknown. Here, to investigate kinetics steps essential for coordination, we observed the movement of kinesin-DNA nanospring complex using a total-internal reflection microscope. When wild-type kinesin and a mutant kinesin which shows prolonged two-head-bound state were attached to both ends of DNA nanospring, wild type kinesin often dissociated from microtubule, while we replaced the mutant kinesin which takes prolong one-head-bound state, both heads dissociated with nearly equal probability. These results suggest that internal load is likely to promote dissociation of kinesin in the one-head-bound state.

3Pos088 INHIBITION OF MITOTIC KINESIN EG5 BY KOLAFLAVANONE

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Mitotic kinesin Eg5 is a validated target in the anti-mitotic therapy for the control of cancer cell proliferation. Here, we report the inhibitory effect of kolaflavanone (KLF), a Garcinia biflavonoid, on the ATPase and microtubule gliding activities of Eg5 in vitro. We also provide evidence of possible mechanism of interaction between the compound and Eg5, as depicted in silico. The results revealed that KLF inhibited the basal and microtubule-activated ATPase activities of Eg5, respectively and suppressed microtubule gliding of Eg5 in vitro. Eg5-KLF model obtained in silico predicted that the biflavonoid is resident within the α2/α3/L5 pocket. KLF binds to Eg5-ADP more tightly than Eg5-ATP. These results suggest KLF as a novel inhibitor of mitotic kinesin Eg5.

3Pos089 F_1-ATPase の軸とシリンダーの結合寿命の測定
Single-molecule pull-out manipulation of the shaft of the rotary motor F_1-ATPase

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Subcomplex of F_1-ATPase is a rotary motor protein in which the central γ-subunit rotates inside the cylinder consisting of $\alpha_3\beta_3$ subunits. To investigate interactions between the γ shaft and the cylinder at the molecular scale, load was imposed on γ through a polystyrene bead by 3-D optical trapping in the direction along which the shaft penetrates the cylinder. Pull-out event was observed under high-load, and thus load-dependency of lifetime of the interaction was estimated. Notably, accumulated counts of lifetime were comprised of fast and slow components. Because the mutant, in which the half of the shaft was deleted, showed only one fast component in the bond lifetime, the slow component is likely due to the native interaction mode held by multiple interfaces.

3Pos090 Does giraffe kinesin move faster than mouse?

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Many neurodegenerative diseases are known to be caused by impaired axonal transport due to decreased velocity of kinesin. For example, point mutations of KIF5A is known to be causative for hereditary spastic paraplegia (HSP), which mainly affects the distal part of the long motor tracts in the spinal cord. The HSP mutations slightly decreased the velocity of KIF5A that would explain why neurons with longest axons are affected. If fast velocity is important for the survival of neurons with long axons, large animals with longer axons would require faster kinesin. Here, we asked a simple question; is kinesin of giraffe faster than small animals such as mice? Our data suggest that KIF5a of large animals with longer axons might have adapted for the longer axonal transport.

3Pos091 Microsecond-resolved observation of F_1-ATPase conformational changes by single molecular fluorescence spectroscopy

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Conformational changes of F_1-ATPase induced by ATP hydrolysis are too fast to be detected by conventional single-molecule fluorescence measurements whose time resolution are milliseconds. We developed the original method of single-molecule Foerster resonance energy transfer (FRET) measurements, and tracked the time evolutions of single-molecules FRET efficiencies with 10-μs resolution. Despite enough time resolution, we could not detect significant time dependent changes in the FRET efficiencies of the sample at 1 mM ATP. On the other hand, at 1 mM of ATPγS, F_1-ATPase exhibited two FRET efficiencies corresponding to two conformations. The results might suggest that the conformational changes of F_1-ATPase upon the phosphate releases occur faster than 10 μs.

3Pos092 遺伝子工学的に人工的に設計したモータタンパク質収縮ネットワークの性能向上
Improvement of a genetically-engineered microtubule contractile protein network

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We have developed a microtubule (MT) network which contracts similarly to muscle fibers when illuminated with UV light. A fusion protein of calmodulin and light meromyosin (CaMLMM) is mixed with a fusion protein of kinesin and calmodulin binding domain such that a complex aggregating MTs is formed upon the release of Ca2+ ions. The force generated, as measured by a pair of PDMS levers, is ~2 μN. The fragility of MT fibers has been identified as the main obstacle for improving the contractile force. To strengthen those fibers, depletion force was generated via addition of polyethylene glycol (PEG), and protein linkers connecting MT filaments have been introduced. The two methods have increased the contractile force generated by 1.5 and 2.9 fold respectively.

3Pos093 暗視野顕微鏡を用いた微小管混雑時における細胞質ダイニンのステップの高時間分解能観察
Cytoplasmic dynein stepping on crowded microtubules resolved using dark-field imaging with high spatio-temporal resolution

Yusuke Kumagai[1], Keitaro Shibata[2], Ken'ya Furuta[2], Hajime Honda[1], Hiroaki Kojima[2] ([1]*Dep. Bioeng., Nagaoka Univ.*, [2]*Adv. ICT Res. Ins., NICT*)

Cytoplasmic dynein and kinesin are motor proteins that move on microtubules (MTs) for intracellular transport, organelle localization and mitosis. Kinesin steps on a single protofilament toward the plus end of the MT, whereas dynein steps on multiple protofilaments toward the minus end of the MT. Previously, we have reported that dynein accelerated on MTs crowded with kinesin, although the acceleration mechanism was unclear. To revel the mechanism, stepping analysis of dynein with high spatio-temporal resolution is essential. In this study, the stepping of dynein attached with gold colloid was observed under a dark-field microscope with high spatio-temporal resolution. We observed how dynein side-steps and bypasses kinesin obstacles on MTs heavily crowded with kinesin.

3Pos094 共通祖先型 F_1-ATPase の一分子回転解析
Reconstruction and Characterization of Ancestral F_1-ATPase

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F_1-ATPase is a rotary motor protein driven by ATP hydrolysis. So far, we revealed that F_1-ATPases from *Bacillus* PS3 (TF_1), bovine mitochondria (bMF_1) and *Paracoccus denitrificans* (PdF_1: a member of the α-proteobacteria) have different reaction schemes. Although phylogenetic analysis suggested that α-proteobacteria is an ancestor of mitochondria, reaction schemes between bMF_1 and PdF_1 are very different. In this study, we resurrected common ancestor of F_1-ATPase and observed the rotation of ancestral F_1-ATPase by single-molecule rotation assay in order to understand what makes these differences of rotation schemes among species. We obtained $\alpha_3\beta_3\gamma$ subcomplex of common ancestral F_1-ATPase of mitochondria/α-proteobacteria. Now we are characterizing its rotational behavior.

3Pos095 クラミドモナス軸糸ダイニン集合体の外部負荷に対する応答を測定する
Measuring mechanical responses of Chlamydomonas axonemal dynein arrays to external load

Misaki Sagawa[1], Akane Furuta[2], Hiroaki Kojima[2], Kazuhiro Oiwa[1,2], Ken'ya Furuta[2] ([1]*Grad. Sch. of Life Science, Univ. Hyogo*, [2]*Adv. ICT Res. Inst., NICT*)

Ciliary and flagellar beating is a consequence of sliding motion of two adjacent doublet microtubules (MTs) that are crosslinked by dyneins arranged in linear arrays along MTs. The continuous beating requires a proper switching of dynein activity. This switching is operated on the time scale of milliseconds, leading us to suppose that external load rather than enzymatic reaction triggers on/off switching of dynein activity. To test the load-dependent regulation of dynein activity, we measured the mechanical response of dynein arrays that were prepared by mixing crude axonemal extracts with polymerized MTs. The measurement was performed by dragging a MT along the dynein arrays with an optical trap. Our study would help understand how dynein activities are coordinated.

3Pos096 (3SHA-4) 過渡的に形成される GPCR ダイマーの研究：細胞内蛍光 1 分子観察によるアプローチ
(3SHA-4) Examining the transiently formed GPCR dimer: an approach by single fluorescent molecule observation in living cells

Rinshi Kasai (*Inst. Front. Life. Med. Sci., Kyoto Univ.*)

Recent studies have shown that G-protein coupled receptors (GPCRs) are in dynamic equilibrium between monomers and dimers in the plasma membrane. However, the functions and properties of GPCR dimer are not well understood. To investigate them, we performed the single fluorescent molecule observation of GPCR and trimeric G-protein in live cells. As a result, we found that dynamic dimer formation of GPCR as well as transient binding of GPCR dimer to trimeric G-protein were modulated by addition of a ligand or an inverse agonist, an inhibitor of constitutive activity. In particular, it was found that trimeric G-protein recruitment to GPCR dimer depends on constitutive activity, suggesting that transient dimer formation is involved in basal signal production.

3Pos097 微小管結合蛋白質を介したアクチンフィラメントと微小管との束化が細胞突起に与える影響
The role of microtubule-associated protein mediated bundle formation between actin filaments and microtubule on cell process formation

Chihiro Doki[1], Masahiro Kuragano[1], Kohei Nishida[1], Shoma Saito[1], Susumu Kotani[2], Kiyotaka Tokuraku[1] ([1]*Grad.Sch.Eng.,Muroran Inst.Tech.*, [2]*Fac. Sci., Kanagawa Univ*)

We recently reported that the microtubule-binding domain (MBD) of microtubule-associated protein (MAP) 4 binds to actin filaments (F-actin), but the physiological roles of the binding remain unknown. In this study, to reveal the physiological function, we examined the behavior of MBD fragments of MAP4, MAP2, and tau when both F-actin and microtubules (MT) coexisted in a sample as well as inside cells. We demonstrated that MT assembly-promoting activity of MAP2 and MAP4 were enhanced in the presence of F-actin and that both F-actin and MT were colocalized with MAP4 in the protrusion of NG108-15 cells. These results implied that MAP2- and MAP4-mediated interaction between F-actin and MT might be involved in the formation and stabilization of cell protrusions.

3Pos098 デスミンとアクチンを包含する液滴のそれらの集合により誘発される変形
Deformations of droplets containing desmin and actin caused by their assembly

Yoshiya Miyasaka, Keigo Murakami, Kuniyuki Hatori (*Grad. Sch. of Sci. and Eng., Yamagata Univ.*)

Intermediate filaments (IFs) are involved in the control of cell migrations. We have shown that the deformation and the protrusion of cell-sized droplets containing desmin IF proteins are induced by desmin assemble forces. Here, we examined whether the co-existence with actin and their states affect the droplet shape. The presence of actin decreased the protrusion rate, whereas increased the deformation rate. Desmin and actin were co-localized. The assembly of desmin was significant for the occurrence of protrusion and co-localization with actin, whereas actin state did not affect colocalization. These results indicate that desmin assembly has a potential to drive the cell-like deformations and determines the co-localization between desmin and actin filaments.

3Pos099 S1P 修飾弾性率可変ゲルを用いた Muse 細胞ホーミング及び力学場応答性の解析
Homing and mechano-response of Muse cells analyzed on S1P-modified hydrogel with tunable elasticity

Lei Guo[1], Yukie Tsuji[2], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

Muse cells are new pluripotent stem cells found in 2010 in adult human mesenchymal stem cell (MSC) population, which is known as repair stem cells in recent years. Tissue repair of Muse cells is performed by preferential homing and spontaneous differentiation responding to the specific conditions of injured tissue, so-called “logics of field”. Although sphingosine 1-phosphate (S1P) has been speculated to contribute to these behaviors, little is known about the details. In this study, employing the S1P-modified hydrogel with tunable elasticity, we have analyzed the effect of S1P and matrix stiffness on the behavior of Muse cells. It was found that S1P immobilized on the hydrogel surface specifically capture Muse cells from MSC population.

3Pos100 細胞の突起形成における MAP4 の局在と機能
Localization and function of microtubule-associated protein (MAP) 4 in cell protrusion formation

Kohei Nishida[1], Masahiro Kuragano[1], Chihiro Doki[1], Susumu Kotani[2], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Eng., Muroran Inst. Tech.*, [2]*Fac. Sci., Kanagawa Univ.*)

We recently reported that microtubule-associated protein (MAP) 4 binds not only to microtubules but also to F-actin and MAP4 bound to F-actin enhanced microtubule assembly-promoting activity in vitro. In this study, to clarify the physiological meaning of the activity, we examined the intracellular localization of MAP4 in NG108-15 cells and influence of MAP4 on protrusion formation. NG108-15 cells were transfected with expression plasmid of GFP-MAP4. The cells were fixed, microtubules and F-actin were labeled, and then observed with a confocal microscopy. The results revealed that MAP4 colocalizes with microtubules and F-actin in cells. We also found that overexpression of MAP4 significantly increased the number and the maximum length of cell protrusions.

3Pos101 F-actin に沿った Fimbrin の協同的クラスター形成の方向性
Direction of the cooperative cluster formation of fimbrin along actin filaments

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Cooperative binding between actin-binding proteins (ABPs) and actin filament (F-actin) is attributed to that the binding of the proteins to the filaments induces cooperative conformational changes of neighboring actin subunits. We previously demonstrated that fimbrin, one of the ABPs, formed cooperative clusters along F-actin. However, the mechanism is unclear how the clusters grow on F-actin. In this study, we clarified the dynamics of cooperative binding of fimbrin to F-actin, which were loosely immobilized on positively charged lipid bilayers, by time-lapse observation. The result of kymograph analysis of the time-lapse images suggested that a large number of clusters grow in one direction (87.4% , n=87).

3Pos102 非凍結温度において氷結合タンパク質は線虫の低温耐性を改善する
Ice-Binding Proteins Improves the Survival Rate of *Caenorhabditis elegans* at Non-freezing Temperature

Masahiro Kuramochi[1,2,3], Geikaku Tou[1], Chiaki Takanashi[1], Motomichi Doi[3], Kazuhiro Mio[2], Sakae Tsuda[4], C. Yuji Sasaki[1,2] ([1]*University of Tokyo*, [2]*AIST-UTokyo OIL*, [3]*Biomedical R.I., AIST*, [4]*Bioproduct R.I., AIST*)

Ice-binding proteins (IBPs) inhibit the ice-crystal growth at freezing temperatures. In addition to this function, IBPs are also thought to stabilize the cell membrane at non-freezing temperatures. Recently, we reported that the expression of IBPs in *Caenorhabditis elegans* improves the survival rate upon cold shock and during freezing (Kuramochi et al., Sci. Rep., 2019). However, the mechanisms of IBP effects on living animals at non-freezing temperatures are unclear. Using transgenic worms expressing IBP, we show that IBP improve the survival rate at non-freezing temperatures. In this session, we discuss that whether adsorption site and thermal hysteresis in antifreeze activity are meaningful factor for non-freezing function *in vivo*.

3Pos103 フェムト秒レーザー誘起衝撃力と反射干渉顕微鏡による細胞接着強度の定量評価手法の確立
Quantitative evaluation of cell adhesion strength by reflection interference contrast microscopy combined with femtosecond laser impulse

Yukiko Yoshimura, Sohei Yamada, Yoichiroh Hosokawa, Ryohei Yasukuni, Kazunori Okano (*Division of Materials Science, Nara Institute of Science and Technology*)

Cell migration is essential to elucidate mechanisms of cancer invasion and metastasis. The cell migration relates with cell adhesion strength on an extracellular matrix (ECM), because cells on the ECM migrate by adhering cell protrusions at leading edge. In this work, we observed the cell adhesion strength by a reflection interference contrast microscopy (RICM) combined with femtosecond (fs) laser impulse. Focused fs laser pulse in water induces a tensile stress wave to the periphery. This stress wave was applied as impulsive force to remove cells from the ECM. RICM detects reflected light from both a glass substrate and cell membrane. Interference of these reflection gives a contrast depending on a distance between them, thus cell adhesive area is visualized.

3Pos104 アクチンネットワーク上におけるアクチン結合タンパク質の局在形成における自律的制御機構
Self-regulatory mechanisms for the segregation of actin binding proteins on actin network

Yosuke Yamazaki, Taro QP Uyeda (*Dep. Phys., Waseda Univ.*)

Amoeboid cell movement requires the proper segregation of actin binding proteins (ABPs), particularly myosin II and cofilin, but this segregation mechanism remains unclear. Here we show that the ABPs formed cell-sized segregation on the actin network whose density was high enough for each myosin filament to bind multiple actin filaments simultaneously. We also show that the cell-sized segregation was not formed when myosin and cofilin were added at the same time, but it was formed when myosin were added before cofilin. These results suggest following self-regulatory mechanisms of actin/ABP system are important for the segregation: mutually exclusive/cooperative actin binding of the ABPs (Ngo, 2016), filmanent-filament distance and interaction order of ABPs.

3Pos105 iPS 細胞は最適弾性領域に移動し、増殖促進と高質な幹細胞性保持を示す
iPS cells show mechanotactic accumulation, higher proliferation and expression of stemness marker in optimal region of matrix elasticity

Mengfan Wang[1], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu. Univ.*, [2]*IMCE, Kyushu Univ.*)

Feeder-free culture of iPS cells on hydrogel substrate has been required for stable and easier supply of them for further clinical applications. One of the problems with such culture system is that iPS cells undergo apoptosis not only due to the loss of cell-cell adhesion but also depending on the quality and strength of cell-substrate adhesion. To establish the optimized physicochemical conditions for the culture of iPS cells on hydrogels, we have scrutinized the effect of stiffness of the hydrogel surfaces on behaviors of iPS cells using microelastically-patterned gelatin gel modified with laminin. We found that iPS cells migrate to the region with optimal stiffness with mechanotactic manner, and exhibit enhanced proliferation and highest expression of Oct4 at the region.

3Pos106 分化フラストレート MSC における APC 発現調節の上流機構の検証
Investigation of upstream regulatory factors of APC expression in the MSCs in frustrated differentiation

Misaki Kaneshiro[1], Thasaneeya Kuboki[2], Satoru Kidoaki[2] ([1]*Grad. Sch. Eng., Kyushu Univ.*, [2]*IMCE, Kyushu Univ.*)

Keeping stemness of mesenchymal stem cells (MSCs) is critical for its quality assurance in medical application. So far, we have found that as MSCs are cultured nomadically on a microelastically-patterned gels to inhibit lineage bias, APC, a key molecule in Wnt signaling, is markedly upregulated. In this study, to seek a factor for stemness maintenance of MSCs, we tried to understand the upstream mechanism of APC expression in such MSCs which we have referred to be in “frustrated differentiation”. Knockdown analysis of potential upstream factors for APC suggested that nomadic migration of MSCs between different regions of elasticity provides mechanical fluctuation of nucleus through cytoskeletal linkage then modulates the gene expression of APC.

3Pos107 Ca^{2+} 存在下/非存在下における mbo1（後退運動変異株）の鞭毛波形
The flagellar waveforms of *mbo1*, a mutant moving backward only, in the presence/absence of Ca^{2+}

Hitoshi Sakakibara, Hiroaki Kojima, Kazuhiro Oiwa (*Adv. ICT Res. Inst., NICT*)

Chlamydomonas increases intracellular Ca^{2+} by intense light stimulation. Then it changes the flagellar waveform from asymmetric to symmetric and moves backward. In *Chlamydomonas*, there are mutants, *mbo*1-3, that always move backward. Here, to investigate the function of MBOs in Ca-dependent waveform change, we analyzed the flagellar waveform of *mbo* 1 under $\pm Ca^{2+}$conditions. In the absence of Ca^{2+}, *mbo*1 flagella showed asymmetry waveform more than the symmetrical waveform of wild-type flagella ($+Ca^{2+}$). It was shown that mbo1 flagella retain Ca sensitivity. The addition of Ca^{2+} increased maximum shear amplitude and asymmetry. These suggest that the waveform changes caused by *mbo*1 mutation and the increase of Ca^{2+} concentration are due to another mechanism.

3Pos108 アクチン結合タンパク質 Rng2 がアクトミオシン in vitro 運動を協同的に阻害するメカニズム
The mechanism of cooperative inhibition of actomyosin movement in vitro by the actin binding protein Rng2

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We previously reported that the actomyosin movement in vitro is strongly inhibited by Rng2CHD, the actin-binding domain of a yeast IQGAP protein Rng2. Half inhibition occurred when the molar ratio [actin]/[bound Rng2CHD] is 75, as estimated from Kd of solution assays. Under those conditions, the helical pitch of actin filaments was shortened by 2-4%. Here we confirmed this strong apparent cooperativity by directly measuring the density of actin-bound GFP-Rng2CHD required for motility inhibition. The apparent strong cooperativity could arise from either memory effect accompanied by transient binding of Rng2CHD to different actin subunits, or true long-distance cooperativity. We are preparing Rng2CHD-actin fusion protein to distinguish those two mechanisms.

3Pos109 二種の異なるアクチン結合タンパク質のアクチンへの結合が相互排他的かまたは協調的かを評価する観察系の構築
Establishment of observation system to evaluate whether two different actin binding proteins bind to actin mutually or accommodative

Tenji Yumoto, Taro QP Uyeda (*Dept. Physics, Waseda Univ.*)

Myosin II and cofilin both bind to actin cooperatively, involving conformational change of actin filaments, and these actin bindings are mutually exclusive (Ngo et al., 2016). This suggested actin filaments regulate selective binding of actin binding proteins (ABPs) allosterically through cooperative conformational changes. To generalize and expand this model, we need to examine if actin bindings of other two ABPs are mutually exclusive or accommodative. Therefore, we polymerized normal actin from seed filaments composed of ABP-actin fusion protein. We are examining the affinity of another ABP to the boundary region between the ABP fusion actin seed and the normal actin to evaluate the ABP binding is exclusive or accommodative.

3Pos110 動的な誘引物質濃度勾配における好中球様 HL60 細胞の運動方向決定
Decision making of migratory direction of neutrophil-like HL60 cells in dynamical chemoattractant gradient

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Neutrophils chemotaxis is thought to be dictated by a mechanism that senses difference in the chemoattractant concentration across the cell body. However, concentration differences in space is temporally changing in vivo environment such as inflammation. Therefore, it is important to understand neutrophils' chemotactic ability under dynamic chemoattractant gradient. Based on precise control of chemoattractant gradient, we show that neutrophil-like HL60 cells exhibit forward movement and concomitant Akt-PH and Cdc42-GTP translocation only in the rising phase of a propagating chemoattractant fMLP gradient. Furthermore, we measured transient response of PI3K and Cdc42 to stepwise change in fMLP concentration to dissect the temporal property of leading edge synthesis.

3Pos111 タウ–微小管相互作用の等温滴定熱測定
Isothermal titration calorimetry of tau-microtubule interaction

Junta Kashima, Hiroshi Sakamoto, Junichi Taira, Hideyuki Komatsu (*Biosci. Bioinf., Kyushu Inst. Tech.*)

Microtubule-binding protein tau is intrinsically disordered in solution. However, tau has been reported to form local conformations upon its binding to microtubules. In order to investigate the binding-induced conformational changes and its thermodynamic properties, we have analyzed tau-microtubule interaction by an isothermal titration calorimetry. Tubulin and human tau40 were prepared from porcine brain, and expressed in *E. coli* and purified by using ion-exchange resins, respectively. The calorimetric titration of tau 40 into tubulin was performed under a tubulin-polymerization condition (in the presence of 5 μM taxol at 308 K). The observed thermogram was exothermic, suggesting a binding to tubulin through basic amino acid residues of repeat domain of tau.

3Pos112 Rhodamine-phalloidin と Lifeact-GFP のアクチン結合の相互排他性
Mutual Exclusion of Actin Binding between Rhodamine-phalloidin and Lifeact-GFP

Saku Kijima[2], **Yuuya Aoki**[1], Taro QP Uyeda[1] ([1]*Dept. Physics, Waseda Univ*, [2]*Biopro. Res. Inst., AIST*)

Lifeact is a short peptide derived from yeast actin binding protein (ABP), and is widely used for labeling of F-actin in vivo. Here, we found that lifeact-GFP formed short clusters along F-actin in vitro, suggesting that lifeact cooperatively binds to F-actin. The lifeact-GFP clusters did not colocalize with rhodamine-phalloidin clusters, suggesting that the actin-bindings of these peptides are mutually exclusive even though the binding sites are unlikely to overlap. Similar mutually exclusive actin-bindings have been reported between cofilin and myosin and between phalloidin and cofilin. To reveal the effect of lifeact on the structure of F-actin and on actin binding of other ABPs, we are investigating the interactions between actin bindings of lifeact and other ABPs.

3Pos113 RhPh 染色したアクチンフィラメントの 3 つの蛍光の明滅パターンの解析
Analysis of three distinct blinking patterns of RhPh fluorescence along actin filaments

Kazunori Ono, Ryuichi Kaneda, **Syunsuke Ando**, Koki Arai, Yosuke Yamazaki, Taro QP Uyeda (*Dept. Physics, Waseda Univ*)

We previously reported that labeling with dilute RhPh shows uneven fluorescence intensities along actin filaments, and that the intensities fluctuate over time. Here, we categorized the fluorescence dynamics into three patterns, depending on the cooperativity along the filament axis and the fluctuation kinetics. Staining with dilute Alexa488-Ph did not produce the two patterns involving cooperativity along the filament axis with fast and slow fluctuation kinetics. Because quantum yield of Alexa488, unlike that of Rh, is insensitive to the local environment, we concluded that the two patterns involve cooperative conformational changes of actin protomers that alter the local environment of bound RhPh. Further studies are underway to unveil the underlying mechanisms.

3Pos114 らせん型細菌スピロヘータの遊泳の力と速度の関係
Force-velocity relationship of the spirochete *Leptospira* swimming

Keigo Abe[1], Kyosuke Takeba[2], Shuichi Nakamura[1] ([1]*Grad.Sch.Eng., Tohoku Univ.*, [2]*Life and Env.Sci., Tsukuba Univ.*)

Leptospira is a member of spirochete, which has a short-pitch helical cell body and two flagella (one at each cell-end). The spirochete moves in liquid by rotating the cell body. The motility is thought to be a crucial virulence factor. However, the mechanism of the infection remains unknown. Motility measurements are important to understand the practical role of motility in infection. In this study, we equipped laser-dark-field microscopy (LDM) with optical tweezers to measure force and speed of individual swimming cells. By using this system, we obtained the force-velocity relationship of *Leptospira* swimming in liquids with or without polymer, e.g., methylcellulose. We will discuss the biological significance of the measured swimming force.

3Pos115 Effect of sucrose on the diffusion of proteins tethered in a glass-supported lipid bilayer

Hiromitsu Hariu (*Saitama Univ.*)

It has been reported that some disaccharides such as sucrose and trehalose play a key role in preserving the structure and function of biomembranes under dry conditions. Various studies have examined the effect of such sugars on the structure and dynamics of lipids in lipid membranes. However, the effect of the sugars on the dynamics of proteins peripherally bound on the membrane is not well studied. Here, we performed fluorescence correlation spectroscopy on proteins tethered in a glass-supported lipid bilayer to analyze how the diffusion of the peripheral membrane proteins is modulated by the presence of sucrose. The results suggested that sucrose accumulates on the membrane surface, which drastically changes the protein diffusion.

3Pos116 分子ツールとしての電位依存性ホスファターゼの改良
Improvement of voltage-sensing phosphatase as a molecular tool of phosphoinositide depletion in living cells

Akira Kawanabe[1,2], Natsuki Mizutani[2], Tomoko Yonezawa[2], Yasushi Okamura[2] ([1]*Fac. Med., Kagawa Univ.*, [2]*Grad. Sch. Med., Osaka Univ.*)

Phosphtatidylinositiol-4,5-bisphosphate [PI(4,5)P2] is an important membrane phospholipid for cell signals through controlling membrane protein activity. Thus, useful molecular tools have been developed for estimating PI(4,5)P2 sensitivity of membrane proteins in living cells. Voltage-sensing phosphatase (VSP) has a PI(4,5)P2 phosphatase activity regulated by membrane potential change. This unique function of VSP provides a tool for PI(4,5)P2 depletion. In this study, we attempted to develop enhanced-VSP (eVSP) based on Danio rerio-VSP (Dr-VSP) for broader versatility and easier usage. We achieved the goal with three improvements: enhancement of intrinsic phosphatase activity, visualization of subcellular localization, improvement of membrane localization. COI:No

3Pos117 油中水滴接触膜張力の定量的操作法とチャネル研究への応用
Manipulation and quantitative evaluation of membrane tension during single-channel current recordings in the contact bubble bilayer

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All the membrane proteins are under the effect of the lipid bilayer tension of the membrane. Previously, we have revealed that the membrane tension fine-tuned the open probability of the pH-activated KcsA channel, implying that membrane proteins are generally subject to change their activity under varied membrane tension. To facilitate tension-dependent characterization, we developed a simple method for manipulation and quantitative evaluation of the lipid bilayer tension on the contact bubble bilayer (CBB). The absolute value of the lipid bilayer tension was successfully evaluated by monitoring the bubble forming pressure and geometrical dimensions of bubbles. This method allows simultaneous recordings of the single-channel activity under arbitrary bilayer tension.

3Pos118 細菌機械受容チャネル MscL の脂質膜環境変化に対する応答のシミュレーション研究
Computational Study Focusing on the Response to Changes of Membrane Environment in Gating of the Bacterial Mechanosensitive Channel MscL

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The bacterial mechanosensitive channel MscL is constituted of homopentamer of a subunit with two transmembrane helices. One of the major issues on MscL is to understand the gating mechanism driven by membrane tension. To get insights into the detailed mechanism of the mechano-gating including the effect of changes in membrane environment, we performed MD simulations for opening of MscL embedded in high/low-density membrane with two ratios of mixture of DPPG and POPG. As a result, MscL in the low-density membrane containing DPPG and POPG with mixing ratio of 1:1 showed spontaneously opening without membrane stretch, thus it is suggested that a combination of the 1:1 mixing ratio and low-density can change its mechanical property for easier opening of MscL.

3Pos119 A gold nano-electrode for single channel detection

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We have developed a novel method for measuring single molecule fluorescence simultaneously with single ion-channel current. We reported an apparatus for simultaneous recording of single channel current and single molecule fluorescence by combining the planar bilayer technique and TIRF microscopy (Ide, ChemPhysChem 2010). Here we report a much simpler technique for simultaneous recordings, in which fluorophores are excited by enhanced electric field at the tip of a very fine electrode and ionic current is measured using the same electrode. We will discuss the development of component technologies.

3Pos120 全自動パッチクランプシステムによる、アダプティブコントロールを用いた正確な 50%不活性化状態制御実験の実現
Adaptive voltage control ensures the precise half inactivation application of voltage gated channels on automated patch clamp system

Kazuya Tsurudome (*Sophion Bioscience K.K.*)

Voltage-gated sodium channels have been studied extensively due to their potential as targets for indications such as pain, epilepsy, cardiac and muscle paralysis. Many of the compounds modulating these channels are state-dependent and preferentially bind to the inactivated state of the channel. The new QPatchII is equipped with the possibility to run online adaptive protocols which makes it possible to measure the half-inactivation potential (V½) for each individual cell and this value may subsequently be used in e.g. a preconditioning pulse. Using this adaptive protocol feature we determined IC_{50} values for both the closed and the inactivated state for a set of compounds and we show that the use of individual V½ reduces data variability compared to standard methods.

3Pos121 人工イオンチャネルの分子動力学シミュレーション
All-atom molecular dynamics simulations of artificial ion channels

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Molecular machines work by converting chemical energy into mechanical energy through conformational changes. Recently, developments of artificial molecular machines, termed molecular engine (ME) are progressing. Kinbara et al. have designed an artificial ion channel (KME). KME consists of hydrophobic transmembrane regions and hydrophilic PEG regions, and its function as a channel is controlled by a ligand binding. However, detailed mechanism of KME has not been understood. To design more efficient KME, we carried out all-atom molecular dynamics simulations of KME embedded in lipid bilayer membrane, and elucidate dynamic features. We found that KME tends to create a pore by multimeric assembly in the presence of ligands.

3Pos122 筋小胞体カルシウムポンプの Ca^{2+}結合に及ぼす界面活性剤の効果
Effect of solubilization with a detergent on sarcoplasmic reticulum Ca^{2+}-ATPase

Takashi Daiho, Stefania Danko, Kazuo Yamasaki, Satoshi Yasuda, Hiroshi Suzuki (*Asahikawa Medical Univ.*)

The effects of a detergent LMNG (lauryl maltose neopentyl glycol) on the Ca^{2+}-ATPase reaction were investigated. Solubilization of the enzyme with LMNG did not inactivate the enzyme irreversibly, but strongly inhibited ATPase activity. The rate of EP decomposition was not retarded. The solubilized enzyme showed very low Ca^{2+} affinity for EP formation from Ca^{2+}-unbound state. However, the high affinity binding of the first Ca^{2+} to the enzyme was not disturbed, and formed Ca_1E1 at μM Ca^{2+} range. Therefore, the affinity of second Ca^{2+} binding was reduced. It was shown that two Ca^{2+} ions are bound to the transition state analog of EP formation in the solubilized enzyme. Results indicate that the solubilized enzyme was stabilized at Ca_1E1.

3Pos123 プロトニック有機電極によるミトコンドリアの ATP 合成操作
Control of mitochondrial ATP synthesis with a protonic biotransducer

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Bioprotonic devices directly interface with the H^+ concentration (pH) to facilitate engineered interactions with biochemical processes. Mitochondrial ATP synthesis is one of important biological activities, which is driven by a H^+ gradient across the membrane. Here we develop a H^+ biotransducer that changes the pH in a mitochondrial matrix by controlling the flow of H^+ between a conductive polymer of sulfonated polyaniline and solution. We have successfully modulated the rate of ATP synthesis in mitochondria by altering the solution pH. Also we succeeded in changing the pH at the cytosol in cells. Our H^+ biotransducer provides a new way to monitor and modulate pH dependent biological functions at the interface between the electronic devices and biological materials.

3Pos124 Toward the construction of DNA origami artificial channel with selective transport function

Shoji Iwabuchi[1], Ibuki Kawamata[1], Yuki Suzuki[2], Satoshi Murata[1], M. Shinichiro Nomura[1] ([1]*Grad. Eng., Univ. Tohoku*, [2]*FRIS, Univ. Tohoku*)

Development of an artificial cell or molecular robot which can work in a microenvironment has attracted attention. As their “body”, closed-lipid bilayer membrane called liposome has been adopted, but the membrane inhibits the input of a hydrophilic molecular signal from the environment. In this work, we design an artificial channel composed of DNA origami that enables to transport desired molecules. The DNA origami has a pore with 10 nm in diameter, which is larger than conventional artificial channels, to pass various kinds of molecules. The passage of molecules through the liposomal membrane has been observed by confocal microscopy. By using DNA strands, we have also designed a lid and molecular mesh to control transportation propaties.

3Pos125 上皮成長因子受容体クラスターによる EGF シグナル伝達の調節
Regulation of downstream signaling by clusters of epidermal growth factor receptor

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Epidermal growth factor receptor (EGFR) has been demonstrated by single-molecule analysis that ligand association alters the receptor mobility to effectively form higher-order clusters, which is suggested to act as a platform for downstream signaling and concern with a membrane subdomain. Depletion of membrane cholesterol, a major component of the subdomain, induced the following effects: 1) ligand-induced dimers increased, but higher-order clusters were decreased; 2) phosphorylation of EGFR gained, whereas that of downstream protein, ERK, was reduced; and 3) translocation of Grb2 adaptor protein became less frequent. Therefore, EGFR higher-order clusters are primary in charge of the downstream signaling, different from the dimers responsible for receptor activation.

3Pos126 p52Shc は時間依存的に Grb2 のシグナル伝達ダイナミクスを制御する
p52Shc regulates Grb2 signaling dynamics in a time dependent manner after cell stimulation

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A cytoplasmic protein Grb2 is translocated to the cell membrane by interaction with ErbB receptor and membrane bound p52Shc (SHC) after cell stimulation with ErbB ligands. We measured the translocation dynamics of Grb2 and SHC in MCF7 cells stimulated with heregulin. The translocation of Grb2 was transient though that of SHC was sustained. Knockdown of SHC decreased the Grb2 translocation early after cell stimulation, while co-expression of an SHC mutant that cannot bind with Grb2 induced more sustained Grb2 translocation. These result suggest that SHC positively regulates Grb2 membrane localization early after cell stimulation but negatively regulates later after cell stimulation. We are currently examining the downstream effects of Grb2 signaling dynamics.

3Pos127 Analysis of electrostatic interaction of acidic glycolipid with transmembrane peptide of insulin receptor

Yuka Nimura[1], Kazuya Kabayama[1,2,3], Yuya Asahina[4], Shinya Hanashima[1], Hironobu Hojo[4], Michio Murata[1], Koichi Fukase[1,2,3] ([1]*Grad. Sch. of Sci., Osaka Univ.*, [2]*MS-CORE, PRC, Grad. Sch. of Sci., Osaka Univ.*, [3]*Inst. for Radiation Sciences, Osaka Univ.*, [4]*Inst. for Protein Research, Osaka Univ.*)

It has been suggested that the electrostatic interaction between insulin receptor and ganglioside GM3 reduces insulin signal and causes type 2 diabetes. However, it has not been shown what kind of interaction actually works because of the complexity of the cell membrane. In order to analyze this interaction, we constructed a model system by incorporating the transmembrane peptide of the insulin receptor into liposomes. We synthesized the fluorescently labeled transmembrane peptide of insulin receptor by using the isopeptide method. The synthesized peptides were incorporated into liposomes and observed by 3D imaging. As a result, it became possible to observe the phase state of the whole liposome which was difficult to discriminate by usual 2D imaging.

3Pos128 細胞膜断片ブレブを用いたモデル生体膜への膜タンパク質再構成
Direct reconstitution of membrane proteins from cell membrane blebs into a model biological membrane

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Model biological membranes are useful tools to study the functions of membrane proteins. We develop a strategy to directly reconstitute mammalian membrane proteins from the cell membrane blebs into a model biological membrane to bypass the technically challenging solubilization and purification processes. We expressed GPCR and GPI-AP in CHO cells and produced blebs by chemical induction. By rupturing blebs on the substrate surface, we could form a planar bilayer and observe single molecules of GPCR and GPI-AP. Furthermore, many reconstituted molecules were observed in a nanometric cleft between the substrate and a PDMS elastomer sheet. This methodology should enable to evaluate the physicochemical properties and functions for a wide range of mammalian membrane proteins.

3Pos129 アガロース微細構造を用いた海馬細胞から伸長する神経突起の相互作用の解析
Interactions of neurites elongated from isolated hippocampal cells in agarose width-length-controlled microchannels

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It is important to understand how neurites interact each other during their elongation for comprehension of neuronal network pattern formation in brain. We investigated the interactions of elongating neurites in agarose microchannel patterns fabricated with a focused infrared laser. In the two microchannels crossing perpendicularly, 55% of two neurites from two hippocampal cells crossed, 35% stopped elongation, 10% elongated in parallel; and in three microchannels gathered and diverged in three fork structure, neurites diverged in three forks when the gathered microchannel width was 16 μm, whereas neurites elongated straight parellely in 21 μm. They suggest neurites do not have any strong attractive tendency and showed repulsive attitudes when they packed together.

3Pos130 Implementation of automated driving by deep reinforcement learning on high definition simulator

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To make a better self-diving agent from sensor information, we used AirSim, a car simulator package in realistic environments using a virtual reality engine. Reinforcement learning is a method to acquire suitable behaviors that maximize value through trial and error based on experience. We trained the steering value using deep deterministic policy gradient which is one of the deep reinforcement learning methods to find the best model. We evaluated the quality of automated driving to the preconfigured destination in a city map. Furthermore, we added a regularization term to stabilize learning. Consequently, experiments of 5000 iterations showed a high average accumulated reward by the agent with relatively stable running.

3Pos131 単一の神経突起における伸長特性を測定するためのスポット吸収マイクロニードルを用いた μm 単位の精度のアガロース微細加工技術
Precise μm agarose microfabrication technology with spot absorption microneedle for single neurite elongation property measurement

Haruki Watanabe[1], Yuhei Tanaka[2], Shota Aoki[2], Kenji Simoda[1], Takahito Kikuchi[2], Akihiro Hattori[3,4], Masao Odaka[3,4], Kenji Yasuda[1,2,3,4] ([1]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*, [4]*WASEDA Biosci. Res. Ins. in Singapore (WABIOS)*)

We have developed a μm-resolution agarose microfabrication technology with a microspot absorption microneedle by a focused 1064-nm infrared laser irradiation to melt a portion of agarose layer by its microspot heat. The minimum size of the microchannel width was μm-order with a precise control of infrared laser power. We fabricated a micro tunnel having two widths, 2-μm for first 100-μm in length and 6-μm for latter 100-μm and observed elongation velocity of single neurite from single hippocampal cell. During the first 2-μm width area, the elongation velocity was 0.24 μm/min, however it dropped to 0.15 μm/min in the latter 6-μm area. The result showed the ability of precise μm-resolution width control technology for neurites elongation velocity control.

3Pos132 畳み込みニューラルネットワークを用いた脳波解析手法の実装
Implementation of EEG analysis method with Convolutional Neural Networks

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Brain-Computer Interface (BCI) is a system which conveys brain signals to computer. Using machine learning, BCI is expected to control a prosthesis limb or to type words. Deep Neural Networks have achieved high classification accuracy and become the efficient tools for data analysis. We use the dataset which contains measurements from 64 electrodes placed on subject's scalps which were sampled at 256 Hz for 1 second. Optimizing EEG channel selection and arrangement is important problem. Our method avoids this challenging process and increases the performance of analysis. We generated 2D heatmaps from the normalized EEG data and trained convolutional neural networks with these images. With ResNet 52, our method reaches 86% accuracy on validation set.

3Pos133 リガンド非結合時のオプシンは稀に光活性化したロドプシンと同等の活性を示す
Apo-opsin exists in equilibrium between a predominantly inactive and a rare highly active state

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Apo-opsin in rod cells activates visual transduction with 10^5-10^6-fold lower efficiency than photoactivated rhodopsin. However, the activity of a single apo-opsin molecule has not been directly measured *in situ* electrophysiologically. Here, we report the detection of apo-opsin activity in a mouse rod. Photoresponse-like events were detected even in complete darkness when only a few apo-opsin molecules were produced in rods. Power spectrum analysis showed almost identical activities between apo-opsin and photoactivated rhodopsin. The event rate reverted to the baseline level after removal of apo-opsin, confirming that the events were derived from apo-opsin. We conclude that apo-opsin exists in equilibrium between a predominantly inactive and a rare highly active state.

3Pos134 天然のバクテリアを用いたヘリオロドプシンの機能研究
Function study of heliorhodopsin using native bacteria

Ai Muto, Rei Abe-Yoshizumi, Hideki Kandori (*Nagoya Inst. Tech.*)

People believed that rhodopsins are composed of two families, type-1 (microbial) and type-2 (animal). However, in 2018, a new rhodopsin family, heliorhodopsin (HeR), was reported through functional metagenomics. More than 500 HeRs are found from Eubacteria, Archaea, Eukaryotes and Viruses. HeR has no ion-transport activity, and slow photocycle suggests sensor function. Nevertheless, function of HeR is unknown to date, and one reason is the difficulty to culture HeR-containing native cells. In this study, we found that some HeR-containing bacteria can be cultured, which encourages experimental analysis toward the function of HeR. Our trial using native bacterial cells will be presented.

3Pos135 メラノプシンの光活性化機構
Photoactivation process of Melanopsin

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Unlike the vertebrate visual opsins whose physiologically active state (Meta-II) thermally decays, vertebrate nonvisual opsins, melanopsins (orthologue of invertebrate rhodopsins), have active state (metamelanopsin), which is stable like that of invertebrate rhodopsins. However, metamelanopsin can be converted to extra form by additional irradiation, which is not reported for invertebrate rhodopsins. It suggests that their active states may diversify. Here we studied the photoactivation process of mouse melanopsin by low temperature UV/Vis spectroscopy. The result showed that formation process of metamelanopsin was similar to that of invertebrate visual opsins. The photochemical property of melanopsin will be discussed in comparing with other opsins.

3Pos136 アミノ酸置換による脊椎動物ロドプシンのバイステーブル特性化
Construction of vertebrate rhodopsin with bistable property by a single mutation

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Among various opsins identified so far, vertebrate visual rhodopsin has a unique molecular property as a mono-stable opsin. That is, vertebrate rhodopsin photo-converts to metastable active state which cannot revert to the inactive dark state. This can lead to precise signal transduction with a higher signal-to-noise ratio than bistable opsins which photo-convert between stable inactive and active states. Thus, the elucidation of the difference of mono- and bi-stable opsins is important to understand highly developed visual system in vertebrates. Here, we present experimental evidence that mon-stable rhodopsin can be converted to a bistable opsin by a single mutation. We will discuss the molecular mechanism specific for formation of mono-stable opsins.

3Pos137 高角 X 線散乱法による光活性化ロドプシンの活性構造安定化メカニズムの解析
Stabilization Mechanism of Active Conformation of Photoactivated Rhodopsin Studied by High-Angle X-Ray Scattering

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Among the photoproducts of vertebrate rhodopsin, only metarhodopsin II (Meta-II) preferentially adopts the active structure as a result of rearrangements of transmembrane helices. Light-induced helical rearrangement of rhodopsin was directly monitored by high angle X-ray scattering (HAXS). We found that the change in HAXS curve upon the formation of Meta-II was different from that of acid-induced active opsin (Opsin*). Analysis using the model structures based on the crystal structures of dark state and Meta-II suggested that the outward movement of helix VI takes place in Opsin*. However, the displaced helices III and V around the cytoplasmic ionic lock in Meta-II are restored in Opsin*, which is likely to destabilize the G protein activating conformation.

3Pos138 シアノバクテリオクロム型光受容体のシステイン残基の着脱反応におけるプロトンの役割
Protochromic Absorption Changes in Two-Cys Photocycle of the Blue/Orange Cyanobacteriochrome

Teppei Sato[1,2], Takashi Kikukawa[3], Risako Miyoshi[4], Kosuke Kajimoto[4], Chinatsu Yonekawa[1], Tomotsumi Fujisawa[4], Masashi Unno[4], Toshihiko Eki[1], **Yuu Hirose**[1] ([1]*Toyohashi Univ. of Tech.*, [2]*Nagoya Univ.*, [3]*Hokkaido Univ.*, [4]*Saga Univ.*)

Cyanobacteriochromes (CBCRs) are phytochrome-related photosensors that show diverse spectral sensitivity. They bind a bilin chromophore and undergo the bilin 15Z/15E photoisomerization upon light illumination. CBCR subfamilies absorbing violet-blue light utilize a cysteine residue to form bilin-thiol adduct, but its relationship to the bilin protonation is not fully understood. Oacu6304_2705 protein photoconverts between blue-absorbing 15Z state and orange-absorbing 15E state. We revealed sequential processes for the reverse photocycle of this protein: the 15E-to-15Z photoisomerization, deprotonation, and thiol adduct formation. These results provide new insight into the molecular mechanisms by which CBCRs absorb light in the violet-blue region.

3Pos139 パターン化モデル膜を用いたロドプシンリン酸化とアレスチン結合の解析
Rhodopsin phosphorylation and arrestin binding studied in a patterned model membrane

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The phototransduction cascade initiated by rhodopsin (Rh) is quenched through its phosphorylation (pRh) and Arrestin (Arr) binding. In spite of its functional importance, localization of pRh and pRh-Arr complex remained unexplored. We recently observed that oligomerized Rh and pRh-Arr complex localize in the central and peripheral regions of the disk membrane, respectively. For elucidating the mechanism of the protein localization, we reconstitute Rh into a patterned model membrane composed of polymeric and fluid lipid bilayers, and microscopically study its phosphorylation and Arr binding. The in vitro assays in a reconstituted model system should give insight into the roles of lipid membrane in the protein localization and signal transduction.

3Pos140 長波長光感受性視物質の塩化物イオン結合における Gln114 の役割
Role of Gln114 in chloride binding of long-wavelength-sensitive visual pigment

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Long-wavelength-sensitive opsins (L opsins) are anion-sensitive in nature. The binding of chloride alters their absorption spectra; this is known as chloride effect. Recent ATR-FTIR spectroscopy studies on primate green sensitive opsin (MG) have provided insights into the role of chloride binding in stabilizing the β-sheet at the extracellular loop (ECL2). Here, we find that Q114 which is positioned far from ECL2 is one of crucial residues for the chloride effect. FTIR spectroscopic analyses on both ion-binding- and light-induced structural changes revealed that Q114 contributes to the stability of β-sheet structure indirectly. Together with further mutational analysis, the functional role of chloride binding will be discussed based on the present studies.

3Pos141 Comparative Study for Anion Transport Activity of Anion Channelrhodopsins by Using a Simple pH Electrode Method

Chihiro Kikuchi (*Grad. Sch. Life Sci.*)

Anion channelrhodopsin (ACR) is a member of microbial rhodopsins and passively transports anions upon light illumination. The anion transport activity has been measured by patch-clamp method applied for mammalian cells and Xenopus oocytes. This method has the benefit of being able to measure quantitatively. However, the experimental procedures are often complicated and thus we need to acquire mature technique. Here we developed a simple method for measuring the anion transport activity of ACRs expressed in yeast cells by using pH electrode. Based on this method, we compared the anion transport activity of several ACRs reported to date. We will discuss the advantages and disadvantages of our method and report new findings for the function of ACRs.

3Pos142 (3SDA-3) 生体組織への応用が期待される光感度の高いチャネルロドプシン
(3SDA-3) Novel optogenetics tool: A light-gated cation channel with high-reactivity to weak light

Shoko Hososhima[1], Shunta Shigemura[1], Hideki Kandori[1], Satoshi Tsunoda[1,2] ([1]*Nagoya Institute of Technology*, [2]*JST, PRESTO*)

Optogenetics has revolutionized the study of experimental biology, making it possible to optically manipulate various biological functions in high temporal and high special resolutions. Especially light-gated ion channels such as ChR2 realized optical excitation when expressed in targeted neurons. Here we report electrophysiological properties and optogenetical application of a light-gated cation channel from cryptophyte, GtCCR4. GtCCR4 exhibits powerful channel activity with a high light sensitivity compared to ChR2. When GtCCR4 was expressed in cultured cortical neurons, successful neuronal firing was observed even by weaker light than required for ChR2 excitation without loss of temporal resolutions, suggesting a potential of GtCCR4 as an ideal neuronal stimulator.

3Pos143 光依存的にホモオリゴマー化する植物クリプトクロム 2 の分子特性
Molecular properties of light-dependent homo-oligomerizing *At* CRY2

Kazuya Agata[1], Daichi Yamada[2], Hideki Kandori[1] ([1]*Nagoya Inst. Tech., Dept. Life Sci. Appl. Chem.*, [2]*Univ. Hyogo, Dept. Life Sci.*)

Cryptochrome (CRY) is a photosensor protein having FAD as a chromophore. CRY2 from *Arabidopsis thaliana* (*At*CRY2) exhibits light-induced homo-oligomerization and interaction with a signaling protein *in vivo*, which are extensively used for optogenetics. However, little is known about the molecular mechanism, and we like to elucidate the molecular mechanism such as structural changes of *At*CRY2 for complex formation. In this study, we successfully observed homo-oligomerization of purified *At*CRY2 *in vitro* as the increase of scattering in UV-vis measurement. We tried to identify the association number of homo-oligomer by SEC and Native-PAGE. We will discuss the molecular properties of *At*CRY2 based on the present results.

3Pos144 光ジッパーを用いた bZIP 型転写因子の標的配列認識の解析
Analyses of the target sequence recognition of a bZIP factor, using a light-activatable Photozipper

Osamu Hisatomi, Samu Tateyama, Itsuki Kobayashi (*Grad. Sch. of Sci., Osaka Univ.*)

Photozipper (PZ) is a basic/region leucine zipper (bZIP) transcription factor containing a light-oxygen-voltage-sensing (LOV) domain. An Asn residue in the basic region (corresponding to Asn131 of PZ) was suggested to play a role in recognition of the target sequence. Site-directed mutant PZs whose Asn131 (N131) were substituted to Ala or Gln showed almost the same spectroscopic and dimerization properties as wild-type PZ. However, N131 mutants showed lower affinities for the target sequence than wild-type PZ. Quartz crystal microbalance data demonstrated that the N131 substitutions accelerated the dissociation without affecting the association, suggesting that a base-specific interaction of N131 occurred after the association between PZ and DNA.

3Pos145 光制御型 bZIP 転写因子（フォトジッパー）の β シート疎水面の役割
Hydrophobic residues on the β-sheet of a light-activatable bZIP factor, Photozipper

Hiroto Nakajima, Itsuki Kobayashi, Osamu Hisatomi (*Grad.Sch.of Sci.,Osaka Univ.*)

Photozipper (PZ) has a basic leucine zipper (bZIP) domain and a light-oxygen-voltage-sensing (LOV) domain. Blue light-induced dimerization elevates the affinity of PZ for the target DNA. The hydrophobic residues on β-sheet at the opposite side to FMN were suggested to have a role in the conformational stability of LOV domain. We prepared five mutant PZs in which amino acids in the hydrophobic region were replaced. Upon illumination, mutant PZs showed similar absorption changes to PZ. However, we detected certain differences in the hydrodynamic radii and affinity to DNA between PZ and mutants. Our data suggested the hydrophobic residues on the β sheet directly affected the conformational switching of PZ either the dark monomeric state or the light dimeric state.

3Pos146 高速 AFM による DNA 結合光受容タンパク質 Photozipper の 1 分子動態イメージング
Single molecular dynamics imaging of DNA binding photoreceptor protein, Photozipper, by high-speed AFM

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Aureochrome-1 (AUREO1) is a DNA binding protein responsible for the blue light (BL) induced blanching of *Vaucheria frigida*. Photozipper (PZ) contains a DNA binding motif (bZIP) and a light-oxygen-voltage-sensing (LOV) domain of AUREO1. Biochemical and spectroscopic studies showed BL induces the dimerization of monomeric PZ, which subsequently increases the affinity of this transcription factor for its target DNA. However, their molecular mechanism still remains unrevealed. We observed single molecular dynamic process of PZ in physiological buffer solution by high-speed AFM. AFM visualized monomer-dimer transition process of PZ molecules diffusing on AFM stage. Under light illumination, AFM movie showed PZ molecules bound to DNA and stayed on the target sequence.

3Pos147 Theoretical study on molecular mechanism of a light-driven ion transport of Halorhodopsin from Natronomonas pharaonis

Ryo Oyama, Taisuke Hasegawa, Shigehiko Hayashi (*Grad. Sch. Sci., Univ. Kyoto*)

Halorhodopsin from Natronomonas pharaonis functions as an inward light-driven chloride pump and is utilized to silence neurons in optogenetics technique in neuroscience. The chromophore retinal isomerizes from all-trans conformation to 13-cis one upon photoabsorption, and triggers a photocycle during which one chloride ion is transported across the membrane. In this study, we performed QM/MM RWFE-SCF calculations to examine the functional coupling of the structural change of the chromophore isomerizing from all-trans conformation to 13-cis one described at the quantum chemistry level of theory with protein large conformational changes of alternating access for the active transport of the ion described with MD simulations with a MM force field.

3Pos148 ウニ初期胚核内構造の発生に伴う動的変化とその細胞特異性
Dynamic and cell specific changes in intranuclear chromosomal

Yuhei Yasui (*Integrated science for life, Hiroshima Univ*)

Recent imaging study for cell nucleus suggested that various intranuclear activities such as gene expression, DNA replications, and DNA repair are regulated by dynamic changes in chromosomal structures. On the other hand, intranuclear structural dynamics during the embryonic development are still unclear. In this study, we observed cell type and developmental stage dependent intranuclear structures of sea urchin embryo, one of typical model organisms for early embryonic development, using fluorescence in situ hybridization, immunofluorescence, and live imaging. We specifically focused on the formation and positioning of nucleolus, centromere, telomere, histone locus body, and heterochromatin to characterize the developmental stage dependent intranuclear features.

3Pos149 ヌクレオソーム排他的ループ非形成型インスレーター配列（NENLIS）によるインスレーター活性のゲノムワイド解析
Genome-wide analysis of insulator activity by nucleosome exclusive non-looping insulator sequence (NENLIS)

Yudai Hirose[1], Yuki Matsushima[1], Naoaki Sakamoto[2], Akinori Awazu[2] ([1]*Grad. Sch. Sci., Univ. Hiroshima*, [2]*Grad. Sch. Integrated Sci., Univ. Hiroshima*)

Insulator sequence is known to play a role of genome domain boundary. CTCF binding site is one of typical insulators that forms chromatin individual loop domain by binding CTCF and cohesin. Recently, nucleosome exclusive sequences like Poly-dA and Ars-insulator identified in Sea Urchin were reported to show insulator activity. Such sequences were called Nucleosome Exclusive Non Looping Insulator Sequence (NENLIS), and more than 5600 NENLIS candidate regions were estimated also in human genome by using Chip-seq, MNase-seq and Hi-C data. In this study, we analyze NENLIS and epigenome states of the regions surrounding NENLIS of human, fly and other organisms to clarify detail properties and the interspecies conservation of such functional noncoding DNA regions.

3Pos150 X 染色体不活性化を誘導する染色体動態
The dynamics of chromosomes that trigger X chromosome inactivation

Tetsushi Komoto, Hiraku Nishimori, Akinori Awazu (*Integrated Science for Life in Hiroshima university*)

The genomic activities of mammalian X chromosomes (X chrs) are mutually supressed to make only one X chr is active in each cell. Such X chromosome inactivation (XCI) enables X chrs number compensation of gene expression level. In mouse, one of two X chrs is inactivated in inner clump of cells on blastocyst stage. The experiment of mouse ES cell suggested the mutual spatial approach of X chrs is essential for XCI start. However, the mechanism of such mutual approach of X chrs in mouse nucleus that contain 40 chromosomes was still unclear. In this study, we constructed a mathematical model of mouse ES cell nucleus. Each chromosome in nucleus was modeled based on Hi-C data and simulated to unveil the mechanism of mutual approach among X chrs.

3Pos151 Machine learning models for predicting ligand-binding sites using residue-wise features

Masafumi Shionyu, Atsushi Hijikata (*Fac. Biosci., Nagahama Inst. Bio-Sci. Tech.*)

To understand molecular function of uncharacterized proteins, annotation of functional sites is important. Therefore, many methods for predicting ligand-binding sites using atomic level features have been developed so far. Such methods usually need precise atomic coordinates. To predict ligand-binding residues from low-resolution structures, we constructed models with residue-wise features, such as ligand-binding propensity, pocketness, and conservation, using XGBoost algorithm, and trained our model on examples of ligand-binding residues as positive data and randomly selected surface residues as negative data. We will discuss the prediction accuracy of our models when the predicted structures by homology modeling were used as input data.

3Pos152 モノクローナル抗体の詳細と提供情報のデータベース
Database for information of acquirable monoclonal antibody

Hirofumi Suzuki[1], Mika Kaneko[2], Yukinari Kato[2], Kei Yura[1,3] ([1]*Dept. Life Sci. & Med. Bio., Waseda Univ.*, [2]*Grad. Sch. of Med., Tohoku Univ.*, [3]*Sim. Info. Bio., Ochanomizu Univ.*)

In modern bioscience, information-sharing platforms for collaborative research have been getting important. Since a number of research projects require multiple resources and methodologies of which specialty and difficulty continues to increase. For instance, a monoclonal antibody is a must in a wide range of bioscience, while an appropriate construct is hard to obtain. We are developing a database for information of antibodies distributed by producer laboratories or acquirable via distributer companies, and will open them public to researchers in the world. As the initial data, we used information on the Antibody Bank (http://www.med-tohoku-antibody.com/). In the poster session, we will discuss the system, user-interfaces, and data deposition of the database.

3Pos153 強力な順方向遺伝学による生物システム解析 -Vibrio alginolyticus の走性への応用
Biological system analysis by a strong forward genetics -An application to a taxis in a strain Vibrio alginolyticus

Kunio Ihara[1], Kazuma Uesaka[1], Noriko Nishioka[2], Seiji Kojima[2], Michio Homma[2] ([1]*Nagoya University Cent.Gene Res.*, [2]*Nagoya University Grad. School Sci.*)

Recent incredible progress in the sequencing technology accelerates to find the responsible gene(s) for an interesting phenotype in a specific biological phenomenon. As one example, we have determined all mutations of15 chemotaxis mutants of an oceanic poroteobacteria Vibrio alginolyticus, which were originally isolated in 1996 by one of the authors. Out of 15 strains, 13 strains had something mutations in the components of the known taxis system, but the two strains had other mutations in the same gene than the known taxis system. This example suggests the existence of an unknown pathway even in the well-studied bacterial taxis system. We would like to discuss the possibility of a novel pathway.

3Pos154 時点数の少ないトランスクリプトームデータからのネットワーク推定に適した遺伝子グループ化法
Gene grouping strategy for network inference from a small time-series transcriptome data

Kiyohiro Maeda (*Fujifilm Corporation*)

Modeling a gene network from a time-series expression data is a potentially powerful approach for investigating context-dependent gene interactions. However, its application has been limited because the available number of sampling time points is often too small relative to the number of genes to make biologically relevant inference. We propose a gene grouping method in which expression pattern and biological function are combined for categorizing genes in order to reduce the imbalance between the number of genes and that of the sampling points. We applied the method to a transcriptome data on human organogenesis and demonstrated that more biologically plausible results can be obtained by modeling interactions after grouping genes for sparsely sampled time-series data.

3Pos155 生物発光タンパク質およびルシフェリンの獲得進化解析
Evolutionary Analysis of Luciferase, Photoprotein and Luciferin

Misato Funahashi[1], Hirofumi Suzuki[2], **Kei Yura**[1,2] ([1]*Grad. Schl Hum. Sci., Ochanomizu Univ.*, [2]*Schl Adv. Sci. Engng., Waseda Univ.*)

Organisms in 117 different families belonging to 12 different phyla have been known to emit light by chemical reaction. The chemical reaction is catalysed by luciferase or photoprotein. We have started collecting data of luminous organisms, including species, habitats, organelles, proteins, ligands and chemical reactions, and revisited the evolution of luminescent systems in the organisms. The proteins was classified into ten groups, and luciferin into nine. No systematic combination of luciferin and luciferase was found. Two organisms were found to use a protein derived from a common ancestor as an enzyme for the same catalytic reaction with completely different residues involved. These data are in the process of compilation and are going to be on a web database.

3Pos156 二倍体遺伝子発現制御系における集合的メンデル遺伝
Group Mendelian Dominance in Diploid Gene Regulatory Network

Kenji Okubo, Kunihiko Kaneko (*Dep. of Basic Sci., Univ. Tokyo*)

The theory of genetics has been modified with adding the interaction between genes, epistasis, to explain the phenomena which does not follow the classical Mendelian dominance. On the other hand, it has been known that many genes interact each other. In this picture, since phenotype cannot be uniquely explained by one gene, it is not obvious for many species to follow Mendelian dominance. To testify Mendelian dominance in those interaction, a theoretical model of diploid gene regulatory networks is used in this research. In the simulation, Mendelian dominance is achieved as genotype groups. In this presentation, the features and causes of those Group Mendelian Dominance are going to be discussed.

3Pos157 不正確な素子から機能的なネットワークをつくる方法
Cooperative architecture for functional network from sloppy gene expression dynamics

Masayo Inoue[1], Kunihiko Kaneko[2] ([1]*IMS, Meiji Univ.*, [2]*Univ. of Tokyo*)

Gene expression dynamics satisfying given input-output relationships were investigated by evolving the networks for an optimal response. We had reported that three different types of networks and corresponding dynamics evolved depending on the sensitivity of gene expression dynamics. In this presentation, we studied their network properties, e.g. noise robustness or mutational robustness, and found they had different advantages. In particular, the cooperative networks composed with many unreliable genes were more functional in terms of robustness and unforeseen challenge. This is why large complex networks are ubiquitous in real biological systems.

3Pos158 混雑下のナノ～マイクロマシン集団：内部状態と環境の相互干渉
Nano/Micro-machines in the Crowd: Interplay between the Internal State and Surroundings

Yuichi Togashi[1,2] ([1]*RIKEN BDR*, [2]*Grad. Sch. Integ. Sci. Life, Hiroshima Univ.*)

There are a variety of molecular machines in the cell, which itself is a complex machinery at the micrometer scale. Artificial micro/nano-machines inspired by biological systems have been also developed. These machines typically exert cyclic operation, in which function and motion of the machine are coupled. If they work in a crowded environment, they may mechanically interfere with each other. To consider such a situation, we have developed a simple model, in which each machine is represented by a particle whose shape depends on the internal state variable of the machine. Spatiotemporal patterns and phase transitions depending on the density and properties of the machines were observed. We will also discuss possible application for modeling of cell crowds.

3Pos159 3D phase field simulation for macropinocytosis of amoeboid cells

Nen Saito[1], Satoshi Sawai[2] ([1]*Grad. Sch. Sci., Univ. Tokyo*, [2]*Grad. Sch. Arts Sci., Univ. Tokyo*)

Ameboid cell shows drastic deformation in cell shape. Such deformation plays fundamental roles in many biological processes. A remarkable example of the drastic deformation is macropinocytosis, which is actin-dependent endocytosis and defined as the non-specific uptake of the extracellular fluid by internalization of plasma membrane. We introduce a mathematical model based on 3D phase field method, which enables to simulate reaction diffusion on the membrane and membrane deformation simultaneously. Simulation results indicate that simple chemical reactions lead to drastic membrane deformation, which results in the engulfment. This study provides a new insight for macropinocytosis as a self-organization via reaction-diffusion on deformable membrane.

3Pos160 Geometric feature extraction from some gene expression pattern for prediction of atopic dermatitis patients

Takuya Hasebe[1], Masahiro Sugimoto[2], Takanori Sasaki[1] ([1]*Grad. Sch. Adv. Math. Sci., Meiji Univ.*, [2]*RDCMIT, Tokyo Med. Univ.*)

In general, discrimination of a disease by machine learning (ML) is executed based on the differential expression (DE) levels of the specific genes between normal and diseased cells. In this study, we aimed to develop a method to discriminate diseases with accuracy even from the gene group with minor DE levels. Specifically, we made a radar chart by three kinds of DE genes from atopic dermatitis (AD) patients, those are ranked from top 248 to 250 th on the DE levels. Subsequently, the values reflected geometric feature of the chart were applied to the ML techniques. As a result, the prediction accuracy for AD by discriminant analysis was improved more than 20%, suggested that combination of minor DE patterns has information for disease onset.

3Pos161 増殖系と隠れマルコフモデルの対応に基づく学習の考察
A Study on Learning in Growing Population on the Basis of Hidden Markov Model

So Nakashima[1], Tetsuya J. Kobayashi[2] ([1]*grad. school of IS&T, UTokyo*, [2]*IIS, UTokyo*)

Growing populations are considered to employ a mixture strategy of a bet-hedging and an adaptation to survive in a fluctuating environment. To understand such survival strategies, the relationship between fitness and a fixed strategy have been discovered in the last decade. The next challenge to reveal the survival scheme is a study on learning of a strategy. On this line of research, Xue and Leibler proposed a learning method in a specific model. However, its mathematical validity and a general methodology of learning are still open problems. In this work, we study learning of a strategy on the basis of the correspondence between population dynamics and Hidden Markov models. Our result is an indispensable step to understand the variety of learning in living systems.

3Pos162 脳神経系の動的ネットワークモデルにおける自発的階層構造形成
Spontaneous hierarchical structure formation in dynamic network model of cerebral nervous system

Amika Ohara, Hiraku Nishimori, Akinori Awazu (*Dept. of math. and life sci. Hiroshima univ.*)

Living systems contain various networks such as neural networks and gene networks. Dynamics of nodes, inducing biological functions, are encoded by the topological properties of such network. Additionally, these networks involve the plasticity where the topological properties of them change through the development as well as the learning and evolution. Therefore, the study of dynamical networks with plasticity provides rich insights to reveal the universal aspects of biological networks. In this study, we consider a neural network model based on a global coupling map system with the connection change that reflects Spike timing dependent plasticity in the nervous system, and focus on the hierarchical network formation observed in cerebral nervous system.

3Pos163 Fitness response relation of a multitype age-structured population dynamics

Yuki Sughiyama[1], So Nakashima[2], Tetsuya Koabayasi[1] ([1]*IIS, The University of Tokyo*, [2]*Department of Mathematical Informatics, The University of Tokyo*)

Control of the stationary population growth rate is ubiquitous problems in many fields. Theoretical studies to analyze its behavior have been often conducted by differential-equation approaches by focusing on the time-slice distribution of the population. However, these approaches can not be applied to a cell lineage tree data, which describes a growing cell population over hundreds generations. To analyze the lineage tree data, we establish a pathwise approach for a multi-type age-structured population dynamics, by employing the large deviation theory for semi-Markov processes. As a result, we find that the responses of the stationary population growth rate with respect to environmental changes can be calculated by the retrospective tracking on the cell lineage tree.

3Pos164 Simulation and regulation of E.coli which has autonomous diversification ability

Eriko Nakagawa[1], Shotaro Ayukawa[2], Daisuke Kiga[1] ([1]*Department of Electrical Engineering and Bioscience, Waseda University*, [2]*Waseda Research Institute for Science and Engineering, Waseda University*)

Synthetic biology allows understanding for principles of bio-network through construction of artificial gene circuits based on mathematical model. For example, in toggle switch, where two repressors are suppressed mutually, change in promoter strength results in bifurcation between mono- and bi- stability. We have combined the toggle switch and cell-cell communication system to implement the bifurcation depending on increase of communication strength. However, saturation of expression due to nonlinearity prohibited emergence of the other monostable point. Both in modeling and living-cells, here, adjustment of a promoter strength in same network topology created the new opposite monostable point by high concentration of communication molecule.

3Pos165 Probability landscape of coupled epigenetic and genetic network with eddy-like probability currents

Bhaswati Bhattacharyya, Masaki Sasai (*Department of Applied Physics, Nagoya University*)

Epigenetic modifications along with transcription process control gene expression in eukaryotes. This coupled stochastic dynamics is described by a model involving discrete state changes of histones and transcription. In case of self-activating gene expression, dynamical equations of order parameters related to protein copy numbers, histone states and transcription states are obtained. We find multiple basins of attraction appear in the probability landscape, depending on the rates of state changes. The divergence-less probability flux is derived from Fokker-Planck equation, which forms eddy current on the probability landscape. We report presence of multiple eddies corresponding to those basins, and discuss time-scale separation in gene expression dynamics.

3Pos166 ドロップアウトを適用したニューラルネットワークアルゴリズムによる大腸菌の遺伝子制御ネットワークの推定
Inference of gene regulatory network of E.coli by neural network algorithm applied dropout

Yusuke Mizukoshi[1], Masahiro Sugimoto[2], Takanori Sasaki[3] ([1]*Grad. Sch. Adv. Math. Sci., Univ.Meiji*, [2]*RDCMIT, Univ.Tokyo Med*, [3]*Grad. Sch. Adv. Math. Sci., Univ.Meiji*)

Neural network (NN) is one of the variety models to predict gene regulatory networks (GRN). In this study, we aimed to improve the predictive accurary for GRN by NN algorithm introduced “dropout” which randomly invalidates the calculation in the input layer in the process of model learning. In particular, the GRN composed of 10 E.coli genes which reguratory network is known was simulated from those expression data. As a result, the prediction accuracy of the GRN by NN algorithm applied dropout was improved by about 10% comparaed to the normal NN algorithm. This result suggests that the pattern change of the input data by dropout contributes to the improvement of the generalization performance in GRN prediction.

3Pos167 Active Nematic Sperms in Vivo Mouse

Tsuyoshi Hirashima[1], Kyogo Kawaguchi[2], Takuya Omotehara[3], Masahiro Itoh[3], Kenta Ishimoto[4], Michiyuki Matsuda[1,5] ([1]*Grad Sch Med, Kyoto Univ*, [2]*RIKEN*, [3]*Tokyo Medical University*, [4]*Grad Sch Math Sci, The Univ of Tokyo*, [5]*Grad Sch Biostudies, Kyoto Univ*)

Active nematics has been well studied theoretically and numerically on the basis of non-equilibrium statistical physics, but the biological experiments are limited to in vitro such as cytoskeletal polymers and bacterial suspensions. Here we show that in vivo mouse sperms exhibit 3D nematic order with disclination lines in the epididymal tubules. Intravital two-photon imaging found that motile sperms in the tail region of epididymis create dynamical patterns in collectives while immotile sperms in the head region show laminar flow. Moreover, we found that the turbulent flow observed in the tail region of the epididymis requires the pulsatile tube contraction. We discuss a possible physical mechanism underlying in vivo mouse sperm movement.

3Pos168 メチルセルロースとの相分離が誘起する重合過程アクチン線維の紡錘形液晶ドメイン自発形成
Emergence of spindle-shaped namatic domains of filamentous actin during polymerization induced by phase-separation from methylcellulose

Masahito Hayashi[1], Tomoyuki Kaneko[1], Kingo Takiguchi[2] ([1]*LaRC, Frontier Biosci., Hosei Univ.*, [2]*Dept. Biol. Sci., Grad. Sch. Sci., Nagoya Univ.*)

Mixture of filamentous actin (FA) and methyl cellulose (MC) is, as well known, spontaneously separated to two phases. We have investigated the phase-separation process during actin polymerization under microscopes. After the addition of KCl to induce the polymerization, many spindle-shaped domains of FA were emerging in the surrounding MC phase. The FA domains were enlarged with time and fused each other after collision. Each domain had birefringence with slow axis parallel to its longitudinal axis, indicating that actin filaments were aligned on the axis. The density of actin in the FA domain was 700 μM, comparable to that in myofibril. These results revealed that the phase-separation effect induced by MC drives self-organization of nematic liquid crystal of FA.

3Pos169 Traveling band formation of a mutant *Dictyostelium* cell population induced by contact following of locomotion

Masayuki Hayakawa[1], Tetsuya Hiraiwa[2], Yuko Wada[1], Hidekazu Kuwayama[3], Tatsuo Shibata[1] ([1]*Riken BDR*, [2]*Dept. of Phys, Univ. of Tokyo*, [3]*Faculty of Life and Env. Sci., Univ. of Tsukuba*)

The emergence of collective motion induced by an alignment effect between individuals has been observed in systems of a large number of self-propelled particles including animal flock, biopolymers, and cells. While the animals and biopolymers are aligned by spatial cognition and collision, respectively, the alignment mechanism of eukaryotic cells is not fully explored. We investigated how the eukaryotic cells achieve alignment effect using the population of Dictyostelium mutant cells. The population formed traveling bands of cell density, and we found that a contact following of locomotion is essentials for the band formation. We conclude that contact following of locomotion enhanced the alignment and induced the emergence of the traveling band.

3Pos170 高速 AFM による KaiC、KaiB、SasA 間相互作用のリン酸化状態依存性観察
High-Speed AFM observation of phosphorylation state-dependent interactions between KaiC, KaiB and SasA

Kenta Ueda[1], Tetsuya Mori[2], Shogo Sugiyama[3], Takayuki Uchihashi[1], Carl H. Johnson[2] ([1]*Dept. of Sci, Nagoya Univ*, [2]*UnivDept.of Biol.Sci., Vanderbilt Univ*, [3]*Dept. of Phys, Kanazawa*)

Kai proteins (KaiA, KaiB and KaiC) constitute the circadian oscillator in Cyanobacteria in which the phosphorylation state of KaiC is controlled by interaction with KaiA and KaiB with the circadian rhythm in vitro. Recently we succeeded in observing dynamic interaction between Kai proteins at single molecular level using high-speed AFM and found that their affinities strongly dependent on the KaiC phosphorylation state. On the other hand, other proteins related to circadian transcription, such as SasA and CikA also regulate circadian rhythm of Cyanobacteria in vivo. However, it remains unknown how these proteins interact with Kai proteins. In this study we directly observed interaction between KaiC, KaiB and SasA to elucidate how SasA binds to the KaiC in place of KaiB.

3Pos171 Individual cyanobacterial circadian rhythms under chilly conditions

Hiroshi Ito[1], Hinako Maruayama[1], Irina Mihalcescu[2] ([1]*Grad. Sch. Design., Kyushu Univ.*, [2]*LIPhy, Universite Grenoble Alpes*)

Chilly environment can abolish circadian rhythms of poikilotherm or plants. We recently found cyanobacteria also lose its rhythmicity around 20 degree. Moreover, we examined temperature dependency on amplitude and confirmed that Hopf bifurcation occurred in both in vitro Kai oscillator and in vivo population of cyanobacterial cells. Still, the observation of Hopf bifurcation at a cellular population level evokes an important question: was the decrease of amplitude caused by desynchronization among individual cells or loss of rhythmicity of individual cells? This question should be tackled by observation of circadian rhythms at a single cell level. In this presentation, we will report the cold-induced transition of circadian oscillation of individual cells.

3Pos172 多電極システムを用いた心毒性検査のためのハイスループットチャンバーの改良
Improvement of high-throughput chamber for cardio-toxicity testing with multi-electrode array system

Naoki Tadokoro, Tomoyuki Kaneko (*LaRC, FB, Hosei Univ.*)

As drugs always have side effects, toxicity testing is indispensable at the preclinical testing. In particular, cardiotoxicity testing is important because cardiotoxicity induces lethal arrhythmias. The toxicity test takes a huge amount of time and cost, and a method that further improves cost, ethics, and technical aspects is required. We constructed a noninvasive and high-throughput cardio toxicity testing system that could obtain the signals of 64 cardiomyocyte populations at 1 time measurement. Although the signal was small and unsuitable for cardiotoxicity tests, we will improve the signal value to conduct a cardiotoxicity test.

3Pos173 HPD による広視野蛍光 1 分子偏光検出
Wide-field single-molecule fluorescence polarization detection by hybrid photo-detectors (HPDs)

Atsuhito Fukasawa[1], Gaku Nakano[1], Minako Hirano[2], Toru Ide[3], Hiroaki Yokota[2] ([1]*Hamamatsu Photonics K.K.*, [2]*Grad. Sch. Creation Photon Indust.*, [3]*Grad. Sch. Interdisciplinary Sci. and Engineering in Health Sys.*)

We have presented that the Hybrid photo-detector (HPD) (Hamamatsu Photonics) consisting of a photocathode and an avalanche photodiode enables low-background wide-field single-molecule fluorescence detection with high temporal resolution. Here, we demonstrate that HPDs enables simultaneous wide-field single-molecule fluorescence polarization detection. We will present time courses of fluorescence polarization of single-molecule fluorophore whose image was simultaneously monitored by an EM-CCD. The HPD based time-resolved single-molecule fluorescence detection, which is unlike conventional single-molecule detection, provides us with alternative approaches to dynamics of single biomolecules.

3Pos174 蛍光タンパク質の赤外スペクトル測定ー発色団部位の選択的な観測ー
IR spectra of fluorescent proteins -selective measurement of chromospheres-

Hirona Takahashi, Makoto Sakai (*faculty of Science, Okayama University of Science*)

We have developed the new IR spectroscopy for fluorescent molecules in water by combining IR super-resolution technique and transient fluorescence detection. In this study, we succeeded the selective measurement of IR spectra of fluorescent protein chromospheres. In the case of phiYFP, the S/N ratio of IR spectrum was quite high although the concentration of the phiYFP aqueous solution was ~10^{-5} mol dm^{-3}. Because no peak appeared in the amide I and II around 1600 cm^{-1} region, it is concluded that the selective IR measurement of fluorescent protein chromosphere was succeeded. The results for other fluorescent proteins such as TurboGFP and eCFP will be discussed in the presentation.

3Pos175 蛍光計測技術を用いた高分子クラウディングが与える相互作用の評価
Elucidation of the Effect of Macromolecular Crowding to Molecular Interactions using Fluorescence Fluctuation Microscopy Techniques

Fusako Gan[1], Akito Matsui[1], Johtaro Yamamoto[2], Masataka Kinjo[3] ([1]*Grad. Sch. of Life Sci., Hokkaido Univ.*, [2]*Biomed. Res. Inst., AIST*, [3]*Fac. of Adv. Life Sci., Hokkaido Univ.*)

In living cells, molecules are affected by surrounding environment such as other molecules or cytoskeleton, which is Macromolecular crowding (MMC). It is important to elucidate the effect of MMC quantitatively for cell activity. The purposes of this study are to create the guide of model solution which mimics the MMC in living cells and to elucidate MMC effect to the biomolecular interactions. For these purpose, we use Fluorescence Cross Correlation Spectroscopy (FCCS) and Polarization-dependent Fluorescent Correlation Spectroscopy (Pol-FCS). These techniques provide quantitative information about molecular dynamics including molecular interactions, rotational and translational diffusion. Here we demonstrate the effect of MMC under the several conditions of solution.

3Pos176 Examination of backtracking engulfment mechainism in macrophages using on-chip single cell observation assay

Amane Yoshida[1], Yuya Furumoto[1], Toshiki Azuma[1], Takahiro Kitahara[2], Tomoyasu Sakaguchi[2], Masao Odaka[3], Akihiro Hattori[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ.*, [3]*Org. Univ. Res. Initiatives, Waseda Univ.*)

It is important to reveal the driving mechanism of the membrane during phagocytosis in macrophage to understand how the backtracking of engulfment occurs. In our previous study, we observed irregular membrane backtracking after half of the engulfment, which cannot be explained by conventional models of engulfment. To understand this mechanism, we observed phagocytosis of macrophages and tracked the progression of membrane engulfing polystyrene beads by measuring the time course of the central angle of the covered part of the beads. As a result, the angular velocity was accelerated at the latter half. The transitions in phagocytosis might be explained by the balance of actin-driven force and restoring force of the membrane which is suggested in previous studies.

3Pos177 キャピラリー吸引法を用いた単一マクロファージの一連の貪食における余分な体積増加について
Extra volume increase of single macrophage during sequential phagocytosis occurred by using micropipette aspiration measurement assay

Toshiki Azuma[1], Yuya Furumoto[1], Amane Yoshida[1], Masao Odaka[2], Akihiro Hattori[2], Tomoyasu Sakaguchi[3], Takahiro Kitahara[3], Kenji Yasuda[1,2,3] ([1]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*, [2]*Org. Univ. Res. Initiatives, Waseda Univ.*, [3]*Dept. Pure & Appl. Phys., Sch. Adv. Sci. & Eng., Waseda Univ*)

We have developed micropipette aspiration measurement assay to measure accurate volume increase of single macrophage during sequential phagocytosis. We aspirated and held macrophage into micropipette and evaluated its volume increase as its length increase with fixed cross-sectional area of its cylindrical shape of inner diameter of micropipette. When we applied 4.5 μm antibody-coated polystyrene spheres, the volume increases of macrophages were 146% more than the volume of applied polystyrene spheres. This result suggests that volume increase of macrophage's engulfment is not so simple as volume increase of only antigens.

3Pos178 Releasing SecM translation arrest and observing resumed translation using magnetic tweezers

Zhuohao Yang, Ryo Iizuka, Takashi Funatsu (*Grad. Sch. of Pharm. Sci., The Univ. of Tokyo*)

SecM, an E. coli secretion monitor protein, contains an arrest sequence, which interacts with the ribosomal tunnel to arrest its own translation. The arrest is considered to be released by pulling force of Sec apparatus. However, this release process has not been fully substantiated. To explore whether the physical force can release SecM translation arrest, we have developed a single molecule force measurement system using magnetic tweezers. By applying the forces of several pN to the SecM-ribosome-mRNA arrested complexes, we successfully observed the resumed translation of ribosome in real time. Our results indicate that relatively weak physical force is sufficient to release SecM translation arrest and resume its translation.

3Pos179 (3SGA-5) 転写伸長を制御するメディエーターの１分子超解像イメージングによる分子局在と動態の定量解析
(3SGA-5) Molecular localization and dynamics of Mediator regulating transcription elongation using single-molecule and super-resolution microscopy

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Mediator, a transcriptional coactivator, is a multiprotein complex with multiple functions in transcription activation and chromatin structure alteration. Recently, Mediator is suggested to regulate releasing RNA Polymerase II (Pol II) from promoter-proximal pausing. To elucidate the molecular mechanism, we performed FRAP analysis using domain deletion mutants of Mediator subunit MED26. The results supports a model in which Med26 N-terminal domain plays a role in switching the binding of TFIID to super elongation complex (SEC). We further carried out simultaneous super-resolution and single-molecule imaging of MED26 and Pol II. It enables us to visualize and quantify how molecules interacts with the cluster formed by the other molecules at the molecular level.

3Pos180 酸性細胞環境内の超解像イメージング応用に向けた耐酸性可逆的光スイッチング緑色蛍光タンパク質の開発
Acid-tolerant Reversibly Switchable Green Fluorescent Protein for Super-resolution Imaging in Acidic Conditions

Hajime Shinoda[1,2], Kai Lu[3], Ryosuke Nakashima[3], Tetsuichi Wazawa[3], Kosuke Noguchi[2], Tomoki Matsuda[2,3], Takeharu Nagai[2,3] ([1]*CPR, Riken*, [2]*Grad. Sch. Eng., Osaka Univ.*, [3]*ISIR, Osaka Univ.*)

Reversibly switchable fluorescent proteins (RSFPs) are crucial tags for super-resolution observation of protein localization and dynamics inside living cells. However, their usage in acidic conditions (pH 4.5-6.0) has been limited, due to the high fluorescence pKa (~5-6). Here, we show new RSFPs with new photochromic mechanism, whose switching-off is caused by trans-to-cis isomerization of the chromophore hydroxyphenyl ring that accompanies protonation. The fluorescence intensity, off-switching speed, and switching contrast of the RSFPs are only slightly affected by changes in pH between 4.5 and 7.5. Exploiting these properties, we succeeded in high-contrast super-resolution imaging of cellular architectures in acidic conditions.

3Pos181 Direct observation of heterogeneous starvation response and emergence of surviving subpopulation in the clonal microbial population

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Phenotypic heterogeneity is ubiquitous and widely observed from microbial to mammalian cell populations. In particular, non-genetic diversity plays an important role in the survival of microbial populations under stressful conditions. How does the cell-to-cell heterogeneity emerge and affect the fate of individual cells? We combined micro-device and time-lapse microscopy, and continuously observed growth and stress response of thousands of bacterial cells from their birth to death at the single-cell level under starvation. Our results show the amplification dynamics of heterogeneity in the cellular growth rate, and demonstrate how particular cells exhibiting longer survivability emerge in the non-genetic diversity.

3Pos182 ヘアレスマウスにおける皮質拡延性抑制の近赤外無侵襲測定
Noninvasive near-infrared monitoring of intrinsic optical signals caused by high K+-induced cortical spreading depression in hairless mice

Hiro Yamato[1], Takashi Jin[2], **Yasutomo Nomura**[1] ([1]*Grad. Eng. Maebashi Inst. Tech.*, [2]*RIKEN BDR*)

The novel imaging method leaving intact scalp on the skull using the light at 670, 785, and 975 nm was proposed. In this study, we used hairless mice (Hos:HR-1) since the deterioration of image quality was resulted from the hair. Cortical spreading depression was induced by KCl application through small incision and burr hole on the frontal bone. Intrinsic optical signals through the intact scalp in the observation area were detected. Time course of the signal showed a triphasic feature which was consistent with the intrinsic optical signals through the intact skull. Although the intact scalp weakened the amplitudes significantly, e.g., 4.0 from 6.9 at 975 nm, the signals during cortical spreading depression were sufficient to be detected.

3Pos183 1粒子でナノスケール温度計とナノスケール熱源になる蛍光ナノダイヤモンド
Application of individual fluorescent nanodiamond as nanothermometer and nanoheater

Chongxia Zhong[1], Shingo Sotoma[1,2], Yoshie Harada[1,3], Madoka Suzuki[1] ([1]*Institute for Protein Research (IPR), Osaka University*, [2]*Japan Society for the Promotion of Science(JSPS)*, [3]*QIQB, OTRI, Osaka University*)

Controlling local temperature in wet samples by releasing and measuring the flow of heat at nanoscale has been a challenge in fields ranging from biomedicine to material science. Various approaches have been developed for nanoscale thermal sensing, whereas the choices of heat releasing methods are very limited. Usually the released heat needs to be monitored on-site by adopting a separate approach to precisely measure the temperature. Here, we present the use of an individual functionalized fluorescent nanodiamond as a nanothermometer as well as a nanoscale heat source under a fluorescence microscope to quantitatively examine the local thermal properties surrounding the nanoparticle.

3Pos184 粒子フィルター MD シミュレーションによる高速 AFM データからの分子動態推定
Biomolecular dynamics inferred from high-speed AFM data via particle-filter MD simulations

Suguru Kato, Toru Niina, Sotaro Fuchigami, Shoji Takada (*Kyoto University*)

High speed (HS) atomic force microscopy (AFM) is useful for observing the structural dynamics of biomolecules. However, being a scanning experiment, the AFM image has temporal disparity among pixels in each snapshot. To conquer this problem, we employed the particle-filter molecular dynamics (MD) simulation, in which the AFM cantilever probes the surface sequentially pixel by pixel. We tested our scheme in the case of synthetic HS-AFM data. In the test, we utilized a trajectory of nucleosome dynamics as the reference, inferred the structural dynamics from a synthetic HS AFM data generated from the reference trajectory, comparing the inferred dynamics from the ground truth data.

3Pos185 Elucidation of the aggregation of serum amyloid A protein and health diagnosis using a high-throughput screening system

Xuguang Lin[1], Masahiro Kuragano[1], Kenichi Watanabe[2], Kiyotaka Tokuraku[1] ([1]*Dep. of App. Sci. and Eng., Muroran Ins. of Tech.*, [2]*Obihiro Univ. of Agric. Vet. Med.*)

Amyloidosis are characterized by extracellular deposits of amyloid, β sheet-rich fibrils, and classified into more than 30 types in the medical field of both domestic animals and humans. Serum amyloid A (SAA) is one of the cause of Amyloidosis, and the amyloid enhancing factor (AEF) has been found to promote SAA produces more aggregation. However, it is obscure how this process works in mechanism of onset of AA amyloidosis. In this study, we examine the effect of AEF in SAA aggregation using Microliter-Scale High-throughput Screening system with Quantum dots nanoprobes. Here, we demonstrated mouse and feline AEF promoted the aggregation of mouse SAA. Our findings will be helpful to understand the pathogenesis of AA amyloidosis, especially the progression on the disease.

3Pos186 3D time-laps imaging of alpha-synuclein aggregation using quantum-dot nanoprobes

Min Nuo[1], Masahiro Kuragano[1], Q.P Taro Noguchi[2], Kiyotaka Tokuraku[1] ([1]*Dep. of App. Sci.and Eng., Muroran Ins. of Tech.*, [2]*NIT, Miyakonojo College*)

Alpha-synuclein, which is a small protein of amino acid sequence 140, is thought to cause the development of Parkinson's disease. Abnormal accumulation of amyloid fibrils constructed from α-synuclein forms Lewy bodies and transmits them to adjacent neurons, causing the reduction of dopaminergic neurons in the dense part of the substantia nigra in the brain. Recently, we succeeded in direct imaging of amyloid β (Aβ) and tau aggregates with a fluorescence microscope using a quantum dot (QD) nanoprobe, and developed that an automated high-throughput screening system of amyloid aggregation inhibitor using this imaging method. In this study, we tried to visualize aggregation of α-synuclein using QD nanoprobe, and successfully imaged the aggregation as 3D time-laps images.

3Pos187 Three-dimensional analysis for formation process of amyloid β_{42} aggregation using quantum dots nanoprobe

Masahiro Kuragano, Kiyotaka Tokuraku (*Div. of Sust. and Env. Eng., Muroran Inst. of Tech.*)

Alzheimer's disease is a progressive disorder of brain that gradually decreases thinking and memory skills. The amyloid cascade hypothesis is that the neurodegeneration in Alzheimer's disease causes abnormal accumulation of amyloid β (Aβ) in the aged brain. However, the detailed process of the $A\beta_{42}$ aggregation and accumulation is still unclear. Previously, we developed a real time imaging method of $A\beta_{42}$ aggregation using quantum dot nanoprobes in the solution. In this study, we performed observation of $A\beta_{42}$ aggregation with high spatiotemporal resolution using confocal laser microscopy. 3D observation suggested that $A\beta_{42}$ aggregates with a diameter of several μm turned out, which it gathers like a spiral step and exhibited a mesh-like structure.

3Pos188 高速 AFM を用いた HECT 型ユビキチンリガーゼのユビキチン化に伴う構造動態の観察
Observation of the structural dynamics associated with ubiquitination of HECT-type ubiquitin ligase using high-speed AFM

Ikumi Mruo[1], Takahiro Watanabe-Nakayama[2], Toshio Ando[2], Hiroki Konno[2] ([1]*Grad. Sch. of Nat. Sci. & Technol., Kanazawa Univ.*, [2]*WPI Nano Life Sci. Inst. (WPI-NanoLSI), Kanazawa Univ.*)

Ubiquitin (Ub) conjugation to the target protein (ubiquitination) is accomplished through catalytic cascade by Ub-activating enzyme (E1), Ub-conjugating enzyme (E2) and Ub ligase (E3). To reveal the detailed mechanism of Ub transfer among these ubiquitination related enzymes, we previously observed the structural dynamics of HECT domain of E6AP E3 ligase using high-speed atomic force microscopy (HS-AFM). However, by using biochemical methods, we also revealed that HECT domain received only one or two Ub from E2, while full length E6AP E3 ligase received multiple Ub from E2. In this study, to reveal the detailed mechanism of Ub transfer by full length E6AP E3 ligase, we tried to observe structural dynamics of full length E6AP E3 ligase using HS-AFM.

3Pos189 Kinesin transport on microtubules studied by high-speed AFM

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Kinesin is a motor protein that uses microtubules as pathways for intracellular transport. In contrast to dynein, another microtubule associated motor protein, kinesin is usually not able to change tracks, i.e. switch to a neighboring protofilament on its microtubule. This fact makes kinesin especially prone to be influenced by obstacles in its path. High-speed atomic force microscopy (AFM) is a technique that has successfully revealed details about protein dynamics. Here, the feasibility of high-speed AFM to observe kinesin walking on a microtubule will be assessed and the influences of scanning forces on kinesin transport will be evaluated. Furthermore, we present a preliminary study of the effect of missing tubulin dimers in the kinesins' path.

3Pos190 鎖の細胞膜提示システムの構築とその機能解析
Construction and functional analysis of the glycan display system on the cell membrane

Ayane Miura[1], Kazuya Kabayama[1,2,3], Syuto Miyake[1], Hiroki Syomura[1], Yoshiyuki Manabe[1], Toshiyuki Yamaji[4], Kentaro Hanada[4], Koichi Hukase[1,2,3] ([1]*Grad. Sch. of Sci., Osaka Univ.*, [2]*MS-CORE, PRC, Grad. Sch. of Sci., Osaka Univ.*, [3]*Inst. for Radiation Sciences, Osaka Univ.*, [4]*NIID*)

Glycans are deeply involved in biological phenomena by interacting with a lot of molecules on the cell membrane. In vivo, the glycan structures are diverse. Therefore, it is not easy to demonstrate the correlation between the structure and function of these glycans. To solve this problem, we developed a method to display synthetic glycans on the cell surface using the HaloTag strategy. First, we synthesized HaloTag fluorescence ligand conjugated with several glycans, and introduced to membrane proteins. Furthermore, we observed interaction with the specific lectin using these probes. This result showed the introduced synthetic glycans were functioning properly.

3Pos191 分岐鎖アミノ酸に対する遺伝子コード型蛍光バイオセンサー
Genetically encoded fluorescent biosensor for branched-chain amino acids

Hiromi Imamura, Tomoki Yoshida, Hitomi Nakajima, Sena Takahashi, Akira Kakizuka (*Grad. Sch. Biost., Kyoto Univ.*)

Branched-chain amino acids (BCAAs; leucine, isoleucine and valine) are essential for protein synthesis and for regulation of metabolisms. Here, we developed a FRET-based BCAAs biosensor, which was constructed by inserting cyan and yellow fluorescent proteins to a leucine/isoleucine/valine binding protein (LIVBP). Conformational change of the LIVBP portion of the biosensor was induced by binding of BCAA, resulting in the increased FRET efficiency between two fluorescent proteins. The biosensor detected approximately from 0.01 to 1 mM of BCAAs. The biosensor enabled us to visualize both nucleocytosolic and mitochondrial BCAAs levels of individual living cells as FRET signals, providing a way to elucidate spatiotemporal dynamics of BCAAs in living systems.

3Pos192 Odor-evoked responses in mouse whole brain as detected by BOLD-fMRI analyses with periodic stimulation and independent component analysis

Mitsuhiro Takeda, Fuyu Hayashi, Naoya Yuzuriha, Sosuke Yoshinaga, Hiroaki Terasawa (*Kumamoto University, Faculty of Life Sciences*)

Mice perceives a wide variety of odorant substances, which induces the behaviors of the mice. The olfactory perception involves the activations of specific neuronal pathways over the mouse whole brain. To investigate the olfactory pathways, we previously developed a BOLD-fMRI method that uses repetitive odor stimulation and independent component analysis (ICA). So far, the administration of an odorant substance had been performed by manually operating a syringe pump to infuse the saturated vapor of an odorant substance. We here automated the operation of the syringe pump, to ensure the accuracy of stimulation timing and duration. As a consequence of the automation, the detectability of brain responses and reproducibility were significantly improved.

3Pos193 クライオ電子線トモグラフィー法による糸状仮足中のアクチン繊維とファシンの可視化
Visualization of F-Actin with Fascin in Filopodia by electron cryo-tomography

Naoko Kajimura[1], Takuo Yasunaga[2], **Kaoru Mitsuoka**[1] ([1]*Research Center for Ultra-High Voltage EM, Osaka Univ.*, [2]*Grad. Sch. Comp. Sci. Syst. Eng., KIT*)

Fascin is an F-actin-bundling protein that stabilizes cell protrusions, such as filopodia, in migrating and metastatic cells. In the process of neural circuit formation, the growth cone functions as an antenna for detecting axonal guidance factors present around cells. Thus, understanding the interaction mechanism between F-actin and fascin is important. Cryo-electron tomography (cryo-ET) and subtomogram averaging allow visualizing macromolecular complexes in the physiological conditions at high resolution. In this study, we visualized the F-actin with fascin in situ using a neuronal model cell (NG108-15) by cryo-ET and calculated the complex structure of F-actin and fascin by subtomogram averaging using RELION.

3Pos194 Fluorescent Ca2+ indicators for multiplexed super-resolution imaging at nanoscopic cellular domain

Kai Lu, Tomoki Matsuda, Tetsuichi Wazawa, Takeharu Nagai (*ISIR, Osaka University*)

On microscopic level, Ca2+ imaging has greatly boosted biology including neuroscience and cardiobiology. At nanoscale, prospective marriage between super-resolution microscopy and genetically-encoded indicator could make a different impact: By revealing Ca2+ distribution within intracellular nm-scale compartments such as the mitochondria, giving clues to fundamental Ca2+ functions. However, canonical nanoscopy often require transition of fluorophore between on and off states, to break the Abbe's diffraction limit. Here we report two color-variant Ca2+ indicators carrying such photophysical property. The indicators have minimal spectrum overlap and allow multiplexed 2D nanoscopy in live cells. We also expect their application in volumetric nanoscopy in near future.

3Pos195 (3SFA-5) Intracellular delivery of biologics using magnetically-navigated nanocarrier

Yoshihiro Sasaki, Ryosuke Mizuta, Naoya Kinoshita, Kazunari Akiyoshi (*Department of Polymer Chemistry, Graduate School of Engineering, Kyoto University*)

Various cells secrete exosomes for intercellular communication. They carry RNAs a capable of controlling cellular functions and can be used as drug delivery system nanocarriers. There is the current need to further improve the efficiency of exosome uptake into target cells. Here, we prepared a hybrid of exosomes and magnetic nanoparticles, which could be guided to target cells by a magnetic field for efficient uptake. By applying a magnetic field to the hybrid, exosomes were efficiently transferred intol target cells as conformed by confocal laser microscopy. We also found that differentiation of stem cells to neuron-like cells was enhanced by magnetic induction of the exosome-magnetic nanogel hybrid, indicating maintenance of the intrinsic functions of the exosomes.

3Pos196 3D image construction methods for observation of cell with micro manipulation

Masaru Kojima[1], Yuma Takeuchi[1], Yasushi Mae[1], Tatsuo Arai[2,3] ([1]*Grad. Sch. Eng.Sci., Osaka Univ.*, [2]*UEC*, [3]*BIT*)

As a promising approach to elucidation of biological phenomena, "live imaging technology" that observes life phenomena of living cells and tissues have attracted attention. For observation of living cells inside, super-resolution microscopes which can perform various 3D imaging are widely used. However, these microscopic techniques have a problem that the resolution in the Z axis direction is not high. Therefore, in this research, in order to compensate for the low resolution in one direction, a sectional image is acquired while rotating the observation object, and a 3D model is constructed. We show the superiority of the proposed method by comparing the proposed method and conventional method by simulation and experiment.

3Pos197 Distinct morphologies of integrin-targeted peptide co-assemblies in peritumoral space vs physiological ECM

William Cortes, Sona Roy, Sachie Yukawa, **Toshio Sasaki**, Wu Xia, Ye Zhang (*Ye Zhang Unit, Okinawa Institute of Science and Technology*)

The link between ECM structural changes and metastasis is well reported, suggesting manipulation of cell-ECM mechanics as a potential cancer treatment strategy. Due to interest in hydrogels as an ECM mimic, we synthesize a biocompatible peptide and study its gelation after peritumor injection. To elucidate the relationship between cell mechanics and surroundings, we characterize the gel after in-situ self-assembly. CryoTEM and SEM revealed distinct fiber structure at the tumor-gel interface versus formation in 2D cell culture. Viscoelastic spectra measured by AFM show modified fiber-fiber interaction in peritumoral conditions. Combined with its low cell toxicity, we hypothesize these properties are responsible for this compound’s anti-metastatic properties.

3Pos198 環状柔軟多関節 DNA モチーフの自己集合
Self-assembly of a Flexible Multi-joing Ring Motif

Shiyun Liu, Ibuki Kawamata, Shin-ichiro Nomura, Satoshi Murata (*Grad. Sch. Eng., Univ. Tohoku*)

The invention of DNA origami has expanded the geometric complexity and functionality of DNA nanostructures. By using the DNA origami technology, we propose a flexible multi-joint ring motif, which consists of seven edges. The edges are linked by flexible hinges, allowing the motif to take various conformations. The motif can connect with each other by the self-complementary sequences on edges. We can adjust the number of the flexible hinges and the number of the connectable edges, resulting in the programmable self-assembly of the motif. We evaluated the self-assembled structure by TEM observation. The proposed multi-joint ring motif can provide a novel method to create nanostructures of high complexity, which has a potential to make functional molecular device.

3Pos199 オン・チップマルチイメージングフローサイトメトリーでの血中の循環腫瘍細胞の同定のためのサイズ解析
Size distribution analysis of circulating tumor cell clusters in blood using on-chip multi-imaging flow cytometry

Masao Odaka[1], Akihiro Hattori[1], Kenji Yasuda[1,2] ([1]*Org. Univ. Res. Initiatives, Waseda Univ.*, [2]*Dept. Pure & Appl. Phys., Grad. Sch. Adv. Sci. & Eng., Waseda Univ.*)

We report time-course change of the morphological characteristics of circulating tumor cell (CTC) clusters in prostate cancer-implanted rat blood by using an on-chip multi-imaging flow cytometer. 50 μL blood samples were collected from 2 to 11 days after implantation, and both bright field (BF) and fluorescence (FL) cell images were simultaneously acquired and analyzed. Cells having BF area of 90 μm2 or larger increased exponentially in seven days after the implantation up to 1% of whole white blood cells including cell clusters having 150 μm2 or larger area, which was not observed in control blood. These results indicate the increase of irregular cells such as cell clusters in blood after malignant tumor formation were one potential biomarkers of metastasis progress.

3Pos200 アミロイド凝集阻害物質の自動スクリーニングシステムにより見出した高活性天然抽出物の評価
Evaluation of highly active natural extracts found by an automated screening system for amyloid aggregation inhibitors

Rina Sasaki[1], Masahiro Kuragano[1], Kenji Monde[2], Koji Uwai[1], Kiyotaka Tokuraku[1] ([1]*Grad. Sch. Eng., Muroran Inst. Tech.*, [2]*Fac. Adv. Life Sci., Hokkaido Univ.*)

Alzheimer's disease (AD) is the most popular disease in dementia. Recent studies have shown that aggregation and accumulation of amyloid β (Aβ) are involved in the pathogenesis of AD. In this study, we tried to develop an automated real-time microliter-scale high throughput screening (MSHTS) system for Aβ aggregation inhibitors using quantum-dot (QD) nanoprobes. We analyzed 504 crude extracts from plants using this automated system and revealed the relationship of Aβ aggregation inhibitory activities of plant extracts using a plant-based classification. We also evaluated highly active natural extracts found by this system using ThT assay and MTT assay.

3HSPos001 **ハクセンシオマネキの日周期と親愛なる敵効果**
Circadian and Dear Enemy Effect of Uca lactea

Mei Oshikawa, Ayumi Fuse, Rinka Aida (*Miyazaki Kita High School*)

宮崎県加江田川に生息する絶滅危惧種Ⅱ類のハクセンシオマネキ（Uca lactea）は繁殖期に甲羅が白くなり，オスは四種類のウェービングをする。本研究は，雌雄の大きさや個体数，オスの行動に注目して解析を行い，次の結果を得た。①甲羅の大きさで，雌雄共通の５つのグループに分類できる。②干潟の場所で甲羅の大きさによるグループに差はない。③甲羅が大きいほど，メスの比率が減少する。④干潟のメスの増加に合わせてオスも増える。⑤甲羅の大きさは行動に影響しない。⑥求愛シグナルをする個体としない個体では巣穴に長時間入る時間が異なる。⑦求愛シグナルが盛んな時刻でも，広報シグナルは減少しない。⑧求愛する個体とそうでない個体で異なる日周期を持つ。今回の結果は，ハクセンシオマネキの生殖できる適齢期や，生殖行動の時間を推測でき，より詳細な日周期がわかる。この成果がこのハクセンシオマネキの保護に貢献すると期待する。

3HSPos002 **オカダンゴムシの移動と体重減少速度の関係**
Pill-bag's relationship between movement and weight decrease rate

Takato Mera (*Miyazaki Kita High School*)

私たちは，オカダンゴムシの経過時間に伴う体重減少について調査した。その結果，以下のことが明らかとなった。①オカダンゴムシは 15 分間で体重が 1%程度減少する。②15 分間中 5 分以上移動した個体には，平均移動速度と体重減少速度の間に強い負の相関がある。③15 分間中 5 分以上移動した個体には，移動時間と体重減少速度の間に強い負の相関がある。これらの結果より，オカダンゴムシは体重減少速度が速い生物であることがわかった。その理由として，オカダンゴムシは本来頻繁に移動する生物でないと考えられる。また，体重が減りやすい個体ほど平均移動速度が遅く，移動時間が短いことがわかった。その理由として，体重が減りやすい個体は移動速度または移動時間を抑えて消費エネルギーを抑えていると考えられる。これらの研究を重ねていくことで，オカダンゴムシがエネルギーを消費するメカニズムについて明らかにしていきたい。

3HSPos003 **アサリの潜砂率について**
In what condition do the clams under the sand

Syunitiro Maruta, Sayaka Haruyama (*Miyazaki Kita High School*)

天然のアサリの漁獲高が，年々減少しているのにも関わらず，アサリにとって最も適切な環境が明確でない事について興味を持った。そこで，アサリが砂の中に潜る様子に注目して研究した。実験方法は，水温のみを 2℃ずつ変えて比較した対照実験と，砂の粒の大きさのみを変えて比較した対照実験の 2 種類である。なお，1 回の実験に使用するアサリの個体数は 10 個体とし，実験を行う時間は 3 時間とする。今回の研究で 3 つの結果が得られた。①水温のみを変えて実験を行った際，18℃と 20℃の結果は，ほとんど差は見られなかった。②水温を 22℃まで上げると潜砂率が大幅に下がった。③砂の粒のみの大きさを変えて実験を行った際，小さい粒の方の潜砂率の方が高かった。今回の結果から私達は，天然のアサリの漁獲高の向上だけでなく，養殖業への貢献が期待できるのではないかと考える。

3HSPos004 **レゴで昇降機の作製とその工夫**
Fabrication and devising of elevator with LEGO

Kodai Hirota (*Miyazaki Kita High School*)

レゴマインドストームで上下を昇り降り出来たら面白いと思い，昇降機を作る研究を行った。レゴがロープを巻く形をとり巻くことができれば昇り降りすると考えた。まず，レゴの形，ギアの数，歯数とその順序に着目してそれぞれ試行錯誤を行い，次の結果を得た。①動滑車型にすることで重い機体になっても昇ることができた。②ギアは最低 3 つ必要だった。③モーターには一番多い歯数をつけると安定した。その過程でロープのずれが生じたため，ロープを左右から挟み込む形に改良した。その結果，ずれが小さくなった。これまでの結果を用いてレゴを作ると仮説のように動くようになった。ギアの設置はとても有効的だった。それまで期待が重く，昇らなかった物も昇るようになった。この成果がこれからの宇宙エレベーターの研究や物を運ぶ機械作りの発展に生かしていけると期待する。

3HSPos005 **セルロースチューブ内の結晶析出**
Crystal Deposition in Cellulose Tubes

Kazuto Ozeki (*Miyazaki Kita High School*)

私達は，特殊な機器を使わず，セロハン膜チューブを用いて溶媒蒸発法により，種結晶を用いず，1段階の操作で，短期間に粒形の大きな結晶を得ることを目的としました。そこで私達は溶解度に差が少ない NaCl を用いて，次の4つの条件が粒形の大きな結晶の析出に必要だとわかりました。①低温低湿の環境で析出させ，セルロースチューブにものが接触しないようにすること，②直径の大きなチューブを用いて，結晶が成長できる空間を確保すること，③チューブ内を溶液で満たし，チューブの外形がやや扁平な形になるようにすること，④チューブの両端を挟む力の強いクリップで挟むこと。これらの条件に基づいて実験を行うと，粒形の大きな NaCl の結晶を得ることに成功しました。

3HSPos006 **チャコウラナメクジの重力走性と光走性の関係**
Gravitational motility of slugs

Misaki Inoue, Takanori Matsuda (*Miyazaki Kita High School*)

チャコウラナメクジ（Lehmannia valentiana）は北海道と沖縄以外の全国に生息する軟体動物であり，農作物の食害で駆除されている。また広東住血線虫を保持し，実際に死亡例も確認されている。ナメクジは全国に生息するが，あまり姿を見せず，身近に感じる機会が少ない。しかし，ナメクジは雌雄同体であり，私たちの研究対象のチャコウラナメクジが生涯で，200 個もの卵を産卵することから局地的に大量発生しやすい。現在，チャコウラナメクジは生得的行動には負の光走性が報告されているが，重力走性など多くのことで不明な点が多い。私たちは安全な農作物の提供を目指して，重力走性を生かした防除方法を開発するために，チャコウラナメクジの重力走性と角度の関係、光走性と角度の関係について調査した。

3HSPos007 **スクミリンゴ貝の習性**
Investigating the Habits of apple snails

Asuka Onitsuka (*Miyazaki Kita High School*)

特に九州地方に多く生息し要注意外来生物であるスクミリンゴ貝（Pomacea Canaliculata）はイネやイグサ，レンコンなどの幼植物に食害を与える。本研究ではスクミリンゴ貝の習性を基礎研究から見つけることを目的とする。そこで，田んぼを再現し，スクミリンゴ貝を飼育観察した。その結果，スクミリンゴ貝には密集する習性があることや水温が下がると動きが鈍くなると仮説を立て，実験を行った。その結果，①水温が23℃の時レンガの隙間に入り，入らなかったときは水槽の壁際や角に位置していた。②水温が 5℃下がった時は隙間に入った数は減り，スタート地点からの動きがほとんど見られない個体があった。③水温が 23℃，17℃のどちらの場合でも密集していることが多かった。研究の結果から，スクミリンゴ貝の水温の差による行動の違いや密集する習性があること，狭い隙間を好むことが考えられる。この成果がスクミリンゴ貝の駆除に貢献すると期待する。

3HSPos008 **火山灰と植物の関係性**
The effect of volcanic Ash on plants

Yui Nakashima (*Miyazaki Kita High School*)

火山大国日本で過ごす私達は火山灰の影響を多々受けている。本研究では農作物への影響に着目し，被害の軽減を目的に実験を行い，以下の結果を得た。①腐葉土の上に火山灰を被せた場合（0～10 ㎜）では発芽率の変化はなかったが，6～10 ㎜では 0～4 ㎜より成長がみられた。 ②腐葉土と火山灰を混ぜた場合（0～100%）は 70%から発芽率が減少し，0～100%間では発芽後の成長に負の相関がみられた。③火山灰を被せた量と保水率には正の相関がみられた。また，火山灰を葉の表面に降り積もらせ酸素濃度調査を行うと，火山灰ありの方が酸素濃度の変動が小さく，光合成は妨げられたと考えられる。以上から，植物に対する火山灰の影響は様々で，これらのことを考慮した農作物への対処を必要とすることが分かった。今後は火山灰による保水率と発芽率，発芽後の成長との関係性を見つけることが課題である。

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。

THORLABS

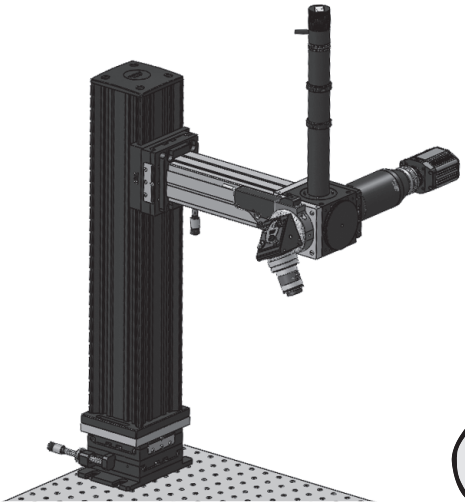
第57回日本生物物理学会年会
バイオフィジックスセミナー

■ プログラム NO. BPS1A
■ 日　時：9 月 24 日 (火) / Sep. 24 (Tue) 11:30 - 12:20
■ 会　場：A 会場 (天蘭) / Room A (TENRAN)

顕微鏡自作支援
ソーラボジャパンの新たな試み
Bring Your Ideas to Life with Thorlabs Parts

勝木 健雄 / PhD. Takeo Katsuki
ソーラボジャパン株式会社 技術部

顕微鏡を作りたい、でもどこから始めればよいかわからない。そのような悩みはありませんか。ソーラボジャパンでは、経験豊富な技術陣が All Ears となって、みなさまの自作顕微鏡のデザインとパーツ選定をサポートします。本セミナーでは、過去の実例を挙げながら、研究者の要望をもとに世界に一つしかないカスタム顕微鏡が生み出される過程をご紹介します。



「広視野回転対物レンズ顕微鏡」設計図
兼子 峰明先生 (理化学研究所 岡野栄之研究室)
定金 理先生 (理化学研究所 山森哲雄研究室)

ソーラボジャパン株式会社

第57回日本生物物理学会年会 バイオフィジクスセミナー

日時 ：9 月 24 日（火）11:30～12:20
会場 ：B 会場（天玉）

細胞動態と遺伝子発現状態を繋ぐ：顕微鏡ライブイメージングと１細胞 RNA シークエンシングの融合と自動化

ご講演 城口克之 先生
理化学研究所　生命機能科学研究センター
RIKEN Center for Biosystems Dynamics Research (BDR)

本研究室では、細胞の動態における分子メカニズムを理解するために、光学顕微鏡で観察した細胞を分取して１細胞網羅的遺伝子発現解析を実現するシステムを開発しています。このシステムを自動化するにあたり、顕微鏡で取得した画像のリアルタイム解析・ステージやフィルターなどの制御などを含むイメージングフローの作製に加え、外付けハードウェアとなる細胞分取装置とのコミュニケーションを行うために、JOBS を有効利用しています。

＊＊＊＊＊＊＊

第57回 日本生物物理学会年会
オプトライン バイオフィジックスセミナー

細胞膜動態のcorrelative imaging

日　時　9月24日（火）　11：30 ～ 12：20

会　場　C 会場（天樹）、Room C （TENJU）

演　者

大場　雄介　先生

北海道大学　医学研究院　細胞生理学教室　教授

細胞膜は脂質二重層や膜タンパク質、糖鎖などで構成され、細胞内外を隔てるバリアである。それは単なる静的構造物ではなく、細胞外物質との相互作用やエンドサイトーシスによる物質の取り込みなどを司り、刻一刻とその形態を変化させる動的素子である。しかしながら、厚さ10 nm以下の細胞膜動態は、100 nm程度のナノスケールで起こる三次元的変化であり、光学顕微鏡の回折限界以下で生じる現象である。我々は、生細胞に特化した高速原子間力顕微鏡（高速AFM）により、これまで誰も見たことがないナノワールド「細胞膜動態」の可視化を行っている。光学顕微鏡では「点」としてしか認識されない微小領域に、複雑な構造とその動きがあることは驚愕に値する。一方、AFMでは細胞膜の機能や分子の特定は不可能で、膜動態の理解には蛍光顕微鏡による同時可視化が必須である。しかし、膜の局所励起により一分子観察を可能とするTIRFやHILOは、大掛かりな光学系のものが多く、振動を嫌うAFMへのアッセンブルは容易ではない。このニーズを満たすために、我々は共同で超小型全反射顕微鏡システムを開発した。そのコンパクトさから、様々なイメージングモダリティーへとの組み合わせの可能性が広がる。本セミナーではこのシステムを紹介し、AFMとの同時可視化例など観察事例を紹介する。

演　者

岩井　亮一　（株式会社オプトライン）

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第57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. BPS1D
◇ 日時: 2019年9月24日(火) 11:30 ～ 12:20
◇ 会場: D会場(天葉)

演題1

「sCMOSカメラの性能はどこまで向上したか?」

"How better is the latest sCMOS camera?"

岡田 康志 先生

理化学研究所・生命機能科学研究センター
細胞極性統御研究チーム
東京大学・大学院理学系研究科・物理学専攻

演題2

「浜松ホトニクスの最新イメージング技術」

- 新製品紹介: 第3世代sCMOSカメラ ORCA-Fusion -

"The latest imaging technology of Hamamatsu Photonics"
- New product: GenⅢ sCMOS camera ORCA-Fusion -

伊東 克秀
浜松ホトニクス株式会社 システム事業部

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第57回日本生物物理学会年会

日本ウォーターズ ランチョンセミナーのご案内

質量分析による蛋白質の高次構造解析
～ネイティブ質量分析と水素重水素交換質量分析～

Mass spectrometry of proteins
– Native mass spectrometry and hydrogen/deuterium exchange mass spectrometry –

大阪大学大学院工学研究科生命先端工学専攻
自然科学研究機構生命創成探究センター（ExCELLS）内山 進 様
Prof. Susumu Uchiyama
Department of Biotechnology, Graduate School of Engineering, Osaka University
Exploratory Research Center on Life and Living Systems, National Institutes of Natural Sciences

日時： **2019年9月24日（火）11:30～12:20**

会場： **E会場**（クリスタル）

質量分析は現代の蛋白質研究において大きな役割を果たしており、プロテオミクスや一次構造解析に加え、高次構造解析においても利用されている。演者らは、ネイティブ質量分析による複合体形成の定量解析を進めてきており、共同利用機関である生命創成探究センター（ExCELLS）ではこれまでに50種類以上の系について複合体形成の測定を行ってきた。

一方、蛋白質の溶液中での相互作用やダイナミクスについて水素重水素交換質量分析（HDX-MS）により解析を行ってきた。セミナーでは、最初に複合体形成についてネイティブ質量分析を用いて解析した複数の結果を紹介し、可能性と限界について説明する。次に、蛋白質-DNA相互作用解析、受容体-低分子リガンド相互作用解析、さらには抗体の凝集につながる部位の特定、をHDX-MSにより行った研究例を紹介し、蛋白質の高次構造解析における質量分析の役割について議論する。

[参考文献]

1. Zhan, Y.Y. et al., 2019, *Nature Communications* 10, 1440.
2. Noda, M. et al., 2019, *J. Pharm. Sci.* in press.
3. Ishii, K. et al., 2018, *BBA General subjects* 1862, 275-286.
4. Kashida, H. et al., 2018, *J. Am. Chem. Soc.* 40, 8456–8462.
5. Uchiyama, S. et al., 2015, *J. Biol. Chem.* 290, 29461-29477.

ヨダカ技研株式会社

日時：9月24日（火）　11:30 ～ 12:20
会場：F 会場（マーブル）

1細胞事業から見つかる「市場が望む研究ニーズと 次世代バイオテクノロジー」

代表取締役 平藤 衛

1細胞（シングルセル）を直接解析したいという研究ニーズが、次世代シーケンサーや質量分析装置の高性能化により、近年高まってきています。

【病気発症のメカニズムを調べたい・病気を診断したい】 がん、糖尿病、アレルギー、アルツハイマー

【寄生虫感染の診断をしたい】 マラリア、フィラリア、アメーバ

【家畜繁殖の効率化や魚類の完全養殖化】 牛、豚、ウナギ、マグロ、ヒラメ

【不妊のメカニズム】 体外受精の助長ではなく、本質的な不妊の改善

【生物の発生メカニズム】 マウス、線虫、ホヤ、プラナリア、トレニア

【再生医療の実用化】 ES、iPS の分化状況、ボディプラン解析

【治療方法の改善】 バイオ医薬、免疫療法、ホルモン療法

上記のような 1 細胞解析に加えて、単離培養（クローニング）の技術向上も求められています。

【環境改善・エネルギー生産に関わる細菌】　セルロース分解、アルコール発酵、汚水処理、土壌改善【抗体生産・ワクチン生産】CHO、ハイブリドーマ、マクロファージ

【有用酵母探索】 医薬品生産、食品生産、ビール、パン、酒

小職が 15年にわたり 1 細胞事業を継続して培ってきた、市場が求めている研究ニーズと 1 細胞研究の展開をご紹介いたします。

〒212-0032　川崎市幸区新川崎7-7 かわさき新産業創造センター（KBIC）
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JST

BINDS 創薬等先端技術支援基盤プラットフォーム
Basis for Supporting Innovative Drug Discovery and Life Science Research

Biophysics Seminar
at the 57th Annual Meeting of Biophysical Society of Japan
11:30-12:20, 24 September (Tue), Room G (Ivory)

1. Recent activities of PDBj and wwPDB
PDBj と wwPDB の最近の活動について

Genji Kurisu

Institute for Protein Research, Osaka University

The PDBj (PDB Japan, https://pdbj.org/) is a core member of the worldwide Protein Data Bank (wwPDB, https://wwpdb.org/) and processes the deposited data from researchers in Asian and Middle-east regions. In order to promote the recent “Data Science”, the wwPDB is introducing several new policies: (i) Collection of ORCID (Open Researcher and Contributor ID: http://orcid.org/) for contact authors has started already and login using ORCID will be implemented in 2019, (ii) Visualization of ligand validation and electron density maps in the wwPDB validation report was improved, (iii) PDBx/mmCIF format became mandatory for MX deposition from July 1st, this year. These issues will be introduced at the Seminar.

2. New tools for editing and annotating structural data

Gert-Jan Bekker

Institute for Protein Research, Osaka University

Ever since the introduction of PDBx/mmCIF, the shadow of death has been looming over the legacy PDB flat-file format. From July 1st 2019, deposition of PDBx/mmCIF formatted files has become mandatory. However, as many users are still used to the legacy flat-file format, we have developed a PDBx/mmCIF editor to help users transition to the new format. The editor doesn’t require any installation and can be used from within a web browser at https://pdbj.org/cif-editor. Users can load local files to edit and save them again, while the tool validates the data with the dictionary. Concurrently, we have also developed a new archive for computationally obtained data, which incorporates the same PDBx/mmCIF editor in addition to our WebGL based molecular viewer Molmil for visualization. The Biological Structure Model Archive is available at https://bsma.pdbj.org, where published entries can be freely downloaded, while new entries can be submitted using users’ ORCID ID.

Protein Data Bank Japan
https://pdbj.org

Institute for Protein Research, Osaka University
3-2 Yamadaoka, Suita, Osaka 565-0871, Japan
TEL: +81-(0)6-6879-4311

dynamic BIOSENSORS

DKSH

第57回日本生物物理学会年会
DKSHジャパン株式会社　バイオフィジックスセミナー

日時：９月24日（火）11:30 – 12:20　会場: H会場（アンバー）

電気的にスイッチングするDNAナノレバーを用いた生物物理学的解析 SwitchSENSE®のご紹介

SwitchSENSE® - Biophysical Analysis with Electrical Switchable DNA nanolevers

坂口　安史（DKSH ジャパン株式会社　科学機器部）

SwitchSENSE はドイツの Dynamic Biosensors 社による、DNA を用いた新しいバイオチップテクノロジーです。このバイオチップの基本構成は、マイクロ流路中の金電極の表面に短い核酸二重鎖（DNA ナノレバー）が固定化されているというユニークなものになっており、従来のバイオセンサーより圧倒的に少ない固定化量で、蛍光による高感度のシグナル検出を行うことができます。

SwitchSENSE は分子間相互作用解析をはじめ様々な生物物理学的な計測を行うプラットフォームとして活用すること可能です。測定モードのひとつ、動的なスイッチングモードでは、固定化された DNA ナノレバーが電場の変化に伴い立ち上がったり倒れたりというスイッチングする挙動をナノレバー先端に結合させた蛍光標識で観測することにより、DNA 先端にコンジュゲートされたタンパク質などの高分子の粒子サイズや構造変化を精度良く計測することができます。（粒子サイズの測定精度は ±0.1 nm~）

また、生体分子間相互作用解析においては、検出限界で約 10 fM の濃度レンジという高感度測定が可能。リアルタイムの結合の速度定数、解離定数を決定できます。その他、タンパク質の熱安定性、化学安定性評価といった測定も実施可能です。また、チップ上で核酸を用いていることから、ポリメラーゼの核酵素カイネティクス解析など、酸関連酵素の様々な解析に容易に応用することが可能です。

今回は、この高感度分子間相互作用・生物物理学的特性解析手法であるSwitchSENSEの原理の概要とアプリケーション例をご紹介いたします。

Selected publications:

Methods 137-145, 118–119 (2017)
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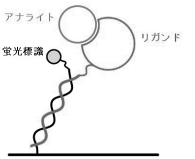

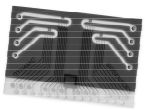

バイオチップ上の DNA ナノレバー

DKSH ジャパン株式会社
テクノロジー事業部門　科学機器部
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第57回日本生物物理学会年会

The 57th Annual Meeting of the Biophysical Society of Japan

プログラムNo: BPS2B

オリンパス株式会社バイオフィジックスセミナー

日　時　9月25日(水) 11:30～12:20

会　場　宮崎シーガイアコンベンションセンター サミットホール 天玉(B会場)

初の国内量産型顕微鏡の開発から100年、さらなる進化を遂げるオリンパスの光学技術

座長：オリンパス株式会社　阿部 勝行

全反射顕微鏡の理想と現実

岡田 康志 先生

東京大学大学院理学系研究科 物理学専攻生物物理学講座 教授

徳永らによる対物レンズ型全反射照明は画期的であった。細胞内での一分子イメージングには、事実上必須の手法である。近年、CMOSカメラの性能が向上し、その広い視野をフルに活かして細胞全体あるいは細胞集団を高精細に撮影することが可能になっている。しかし、対物レンズ型全反射照明で、その視野をフルに活かすことは困難であった。本講演では、光学系の工夫と新しい低収差対物レンズの組合せによる広視野一分子イメージングについて紹介したい。

新世代対物レンズの紹介
～研究の信頼性と効率向上～

オリンパス株式会社　坂倉 正洋

第57回日本生物物理学会年会　バイオフィジックスセミナー

マルバーン・パナリティカル事業部

日時：9 月 25 日　11:30-12:20

場所：C 会場

タンパク質の溶液中安定性の評価方法

―構造安定性・コロイド安定性から凝集定量評価方法まで―

Analyses for prediction of protein stability in solution

-Structural stability, Colloidal stability and quantitative analysis for protein aggregation-

志波　公平
スペクトリス株式会社 マルバーン・パナリティカル事業部
Pharma&Food Sector 事業開発、North Pacific 地域

タンパク質の詳細な構造解析を実施するためには、大量のタンパク質を理想的な分散状態で調製する必要があります。特に X 線結晶構造解析など物性解析を実施するにはそれらの溶液が高濃度に調製する必要もあります。また、近年産業分野でも抗体を治療薬として利用するバイオ医薬品に関しては、産業としてタンパク質を安定的に分散させておく必要があるため、溶液中のタンパク質の安定性を正しく評価する必要があります。

一方で、分散性や安定性の各評価は、本来の研究目的をサポートするための評価であるため、できるだけ本業の負担を減らすこともポイントとなることが考えられるため、サンプル量、必要時間を最小限に留める努力も必要になります。

他方、タンパク質はその大きさが数 nm の高分子であることが知られています。例えば現在治療薬としても使用されている抗体は 150 kDa のヘテロダイマーですが、Hydrodynamic Radius は約 5.5 nm 程度を示します。この大きさは、もちろん分子としての振る舞いも行うのですが、粒子としての振る舞いも無視できない領域に入ってきていることを示します。従いまして、タンパク質の分散性、安定性を評価する際は、高分子骨格自体の安定性だけでなく粒子としての安定性（コロイド的安定性）もまた確認しておく必要があることを意味します。また同時に、分散条件の違いによるタンパク質の凝集量の違いを評価することも効果的な評価手法であると思われます。

本発表では、タンパク質の構造安定性、コロイド安定性評価方法を紹介し、それぞれの特徴について述べる予定です。また、タンパク質の凝集量の定量評価方法も紹介します。

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第 57 回日本生物物理学会年会

サーモフィッシャーサイエンティフィック　バイオフィジックスセミナー

HDX-MS を用いたバイオ医薬品の特性解析

演者　鳥巣　哲生　先生

大阪大学　大学院工学研究科　生命先端工学専攻

日時　9 月 25 日（水）11:30～12:20

会場　D 会場（天葉）

講演要旨

タンパク質を重水素溶液に晒すと、タンパク質分子の水素原子と重水素原子の交換が起こる。重水素への交換速度は水素原子周囲の環境によって変化することから重水素への交換速度を質量分析によって測定することで、タンパク質の立体構造を推定することができる。この構造解析法は水素/重水素交換質量分析（Hydrogen/Deuterium Exchange-Mass Spectrometry, HDX-MS）と呼ばれ、近年タンパク質の高次構造解析や相互作用解析に利用されている。特に、バイオ医薬品の代表例としても知られる抗体については、ヒンジ部の運動性から X 線結晶構造解析など他の手法での解析は困難であることから、HDX-MS による抗体全長での構造解析が効果的である。HDX-MS を用いた抗体全長の構造解析によって、これまで知られていなかった抗体の構造ダイナミクスの変化や相互作用様式が明らかとなってきた。本発表では、HDX-MS を用いたタンパク質の構造解析の概要と抗体医薬品の最新の特性解析事例について紹介する。

第 57 回日本生物物理学会年会　バイオフィジックスセミナー

機能拡張型顕微鏡と赤レーザーアプリケーション

発表日：9 月 25 日 (水)　11:30 – 12:20　会場：E 会場 (CRYSTAL)

シグマ光機株式会社　開発部　井上 裕一

アンコールにお応えしまして、光学機器メーカーのセミナーを開催させて頂きます。今年も著名な先生のご講演はありません。膨大な予算をお持ちの方限定のお話もご用意しません。「予算が膨大でなくても、初心者であっても、学生であっても、独自の顕微鏡や光計測系は始めることができる。そして、メーカーの制約に縛られずに、自由なアップグレードを可能にすることが、おもしろい研究を支える。」そんな製品を目指して新製品を開発しています。

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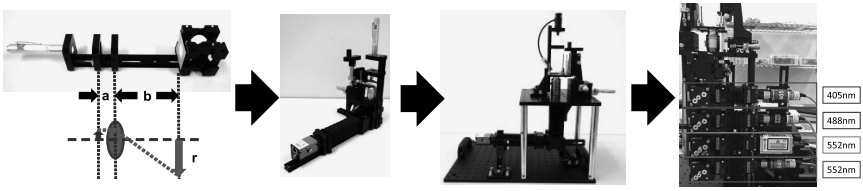

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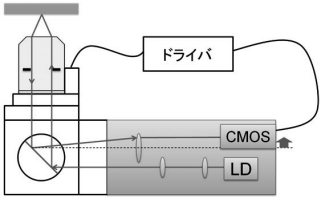

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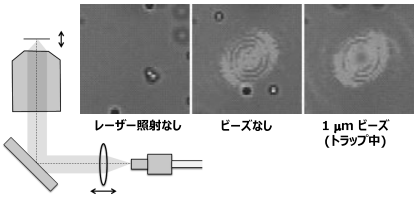

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第 57 回日本生物物理学会年会　ランチョンセミナー

生体物質のナノ構造解析
~ラボ装置の最新技術~

- 日時：2019年9月25日（水）　11:30 ～ 12:20
- 会場：F会場（マーブル）
- 演者：アントンパール・ジャパン
  アプリケーション スペシャリスト 高崎祐一

タンパク質やリン脂質は分子内または分子間の相互作用により特徴的な三次元構造や分子集合体を形成し、生体内の酵素反応・受容体形成において重要な役割を果たします。その機構解明、安定性評価、組成の最適化には、液中における粒子サイズ・形状・内部構造等をナノスケールで正確にとらえる観測手法が必要です。しかし従来の装置は検出器感度やプローブ線源の強度が低く、しばしば非常に希薄に調整される生体試料を正確に測るのが困難でした。

アントンパールの DLS 及び ELS を測定原理とした粒度分布-ゼータ電位計、および小角 X 線散乱装置は非常に希薄な液体試料でも様々な構造情報（粒径分布・粒子形状・粒子内の電子密度分布）を正確かつ迅速に取得することができます。

本発表では、**① Litesizer™ 500 による粒子サイズおよびゼータ電位の測定事例、② SAXSpace によるナノ構造解析事例**をご紹介します。

最近の超遠心分析による生体高分子複合体の溶液物性研究

Modern Analytical Ultracentrifugation for Biological Macromolecular Complexes

内山 進 先生

大阪大学　大学院工学研究科生命先端工学専攻、
自然科学研究機構生命創成探究センター

Dr. Susumu Uchiyama

Department of Biotechnology, Graduate School of Engineering,
Osaka University Exploratory Research Center on Life and Living Systems,
National Institutes of Natural Sciences

超遠心分析の理論と測定原理はスベドベリによって100年近く前に確立され、1980年頃まで生体高分子や合成高分子の溶液中での物性研究において盛んに利用された。1980年～1990年代は電気泳動やクロマトグラフィーにその座を取って代わられたが、測定精度と再現性が高い新型装置が登場し、コンピューターが高速化した2000年以降は、測定データの数値解析を駆使した新しい解析アプローチが可能となり、現在では、生体高分子複合体の溶液物性研究において必須の手法となっている。一昨年にはベックマン・コールター社より最新型の超遠心分析装置OPTIMAがリリースされ、従来と比べ高いスループットで、更に正確な測定が可能となってきている。本セミナーでは、原理を含めた超遠心分析の概要を紹介した後、最近の解析例として、蛋白質の定量的分子間相互作用解析、バイオ医薬品の品質管理、ウイルスの分散度解析、などを他の手法により得た数値との比較も含めつつ紹介する。

Analytical ultracentrifugation (AUC) was invented by Dr. Svedberg more than 100 years ago and extensively used until 1980 for the characterization and understanding of biological and synthetic polymers in solution. Though electrophoresis and chromatography took over the role of AUC in 1980-1990s, after 2000 AUC have become an essential approach for the characterization and understanding of the polymers due to higher precision and reproducibility of the newly released instrument as well as high speed computing ability which enable us to conduct numerical analysis of the AUC data. In addition, using the latest version of AUC instrument, OPTIMA, from Beckman-Coulter, we can now perform highly reliable and high throughput analysis. In the seminar, I' ll introduce principle of AUC and then recent research examples such as quantitative intermolecular interactions, quality control of biopharmaceuticals and size distribution analysis of viruses, together with the comparison of results from AUC with those from other methods.

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第 57 回日本生物物理学会年会
— The Biophysical Society of Japan —

［SCIEX 共催］ ランチョンセミナー
Luncheon Seminars

日時 2019 年 9 月 25 日(水) / Sep.25(Wed) 11:30 ～ 12:20

会場 H 会場（アンバー） Room H（AMBER）

バイオ医薬品の品質管理において、質量分析が担う役割
Role of mass spectrometry in quality control of biopharmaceuticals

株式会社ユー・メディコ
U-Medico INC

野田 勝紀 先生 Masanori Noda

［ 講演要旨 / Abstract ］

近年、抗体医薬品を代表とするバイオ医薬品の研究、開発が盛んに行われている。新しいターゲットに対するバイオ医薬品の開発のみならず、近年では先発品の特許の失効に伴い、バイオ医薬品の後発品であるバイオシミラーの開発も盛んに行われており、この流れはしばらく続くと考えられている。このようなバイオ医薬品の大半の主成分は、組換えタンパク質である。今までの医薬品で主流であった低分子医薬品と異なり、その製造には培養細胞が用いられ、さらに、タンパク質を変性、変質させることなく、長期間保存することが求められている。そのため、培養条件により影響される糖鎖付加の状態などのタンパク質の特性の評価、また保管に伴うタンパク質の化学修飾の評価などを品質管理の観点から行うことが必要である。これらの品質管理は、「最終段階でテストするだけでなくプロセスの段階ごとに確立する必要があり、これにより最終製品の一貫した品質を得ることができる」という考え方に基づいた「Quality by design (QbD)」アプローチに基づいて実施される。そのため、タンパク質の品質を、各プロセスの段階で、多くの分析手法を用いて、確認することが行われてきた。このような分析は、非常に時間と労力を必要とする。そこで、考えられたのが、ペプチドマッピングの利用である。これまで、ペプチドマッピングは、局方における確認試験の一分析として扱われてきたが、近年の質量分析器、また解析手法の発展により、一度のペプチドマッピングにより得ることが可能な情報が非常に多く、定量性に富んだものになったため、マルチ特性分析 (Multi Attribute Methodology：MAM) と呼ばれるペプチドマッピングを元にした分析手法が提唱されることになった。これは、一度のペプチドマッピングの測定で、糖鎖の付加情報、保管などに伴う化学修飾、ジスルフィド結合の同定などの情報を得ることが可能である方法である。本講演においては、こちらの MAM 分析を中心に、バイオ医薬品の品質管理における質量分析の役割についてお伝えしたい。

In recent years, research and development of biopharmaceuticals as typified by antibody drugs have been actively conducted. In addition to the development of new biopharmaceuticals, development of biosimilars, which is a generic product of biopharmaceuticals, has been actively carried out with the expiration of patent of the original product in recent years. Therefore, this trend is expected to continue for a while. The major component of most of such biopharmaceuticals is recombinant protein. Unlike low-molecular-weight pharmaceuticals, which have been the mainstream in medicine until now, cultured cells are used for their production, and furthermore, long-term storage of proteins without being denatured or altered is required. Therefore, it is necessary to evaluate the characteristics of the protein such as the state of glycosylation, which is affected by the culture conditions, and the evaluation of the chemical modification of the protein during storage, from the viewpoint of quality control. These quality controls are based on the idea that “the quality must be established not only at the final stage but also at each stage of the process, thereby obtaining consistent quality of the final product” “Quality by design (QbD)” approach. Therefore, it has been performed to confirm the quality of protein at each process step using many analysis methods. Such analysis is very time consuming and labor intensive. Instead, the use of peptide mapping is being considered. So far, peptide mapping has been treated as an analysis of classical identification tests, but with the recent development of mass spectrometers and analysis methods, there is a great deal of information that can be obtained by single measurement of peptide mapping. Therefore, an analytical method based on peptide mapping called “Multi Attribute Methodology (MAM)” has been proposed. This is a method by which it is possible to obtain additional information on glycosylation, chemical modifications associated with storage, identification of disulfide bonds from one measurement of peptide mapping. In this talk, I would like to talk about the role of mass spectrometry in the quality control of biopharmaceuticals, focusing on this MAM analysis.

第57回日本生物物理学会年会　バイオフィジックスセミナー

マルバーン・パナリティカル事業部

日時：9 月 26 日 11:30-12:20

場所：C 会場

溶液中の高分子 / 粒子個数濃度を算出する技術の紹介

―多角度動的光散乱法 (MADLS) を中心に―

Analysis for concentration of polymer/particle in solution

-A new technique “Multi Angle Dynamic Light Scattering-

志波　公平

スペクトリス株式会社 マルバーン・パナリティカル事業部

Pharma&Food Sector 事業開発、North Pacific 地域

ナノ粒子の製造技術は日々進歩を遂げており、合成・制御できるナノ粒子のサイズはどんどん小さくなってきています。特にフローサイトメトリー法など光学的に検出する量子ドットなどはシングルナノサイズがほとんどであり、物性としても高分子と多くの共通点を有していることがわかります。

一方、溶液中のナノ粒子の定量評価は各実験を進める上で重要な役割を示しますが、そのサイズ領域が高分子のそれと近づくことで困難になったものも存在すると思われます。そのひとつが個数濃度ではないかと思います。そもそも、粒度分布や個数分布、個数濃度の概念は、過去からは「ふるい」技術によって多くを議論されてきたため、その後の粒度分布測定装置にも「重量基準」の概念が残ってきました。そのため、規定のサイズの分布とその重量、そしてそこから計算された実際の粒子個数（個数濃度）が産業的に重要な位置づけとなっている現状があります。

今回紹介する多角度動的光散乱（Multi Angle Dynamic Light Scattering; MADLS）法は、動的光散乱（DLS）法をベースとした技術で、DLS 法によるダイナミックレンジにおける粒子（高分子）個数濃度を計測する技術です。本来光散乱強度は、散乱体積と屈折率によって定まりますので、サイズがわかればそこから個数濃度を算出することが可能でした。特に 0.1 µm 以上の、角度依存を持つ領域を、多点の散乱強度およびサイズ情報（DLS）を獲得することによって DLS 計測可能領域において粒子個数濃度を算出できるようにした技術になります。

本発表では、特にナノレンジにおける粒子濃度を計測するいくつかの技術を紹介し、それぞれの特徴と注意点について触れる予定です。

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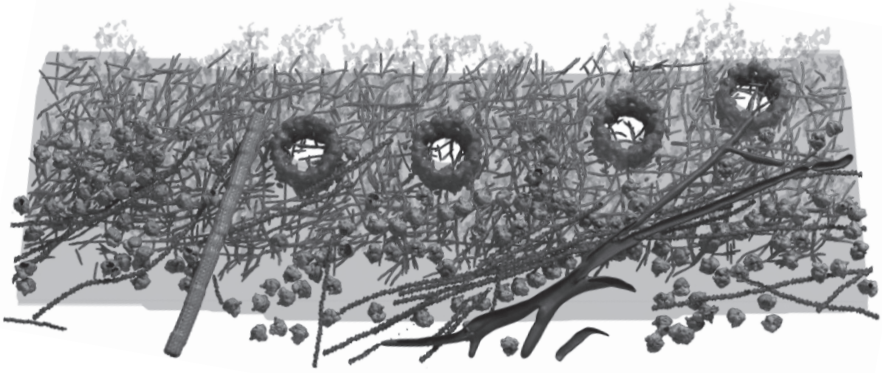
BPS3D　サーモフィッシャーサイエンティフィックグループ
日本エフイー・アイ株式会社ランチョンセミナー
クライオ電子線トモグラフィー法による細胞内立体構造解析への展開



Mahamid et al., Science. 2016 Feb 26;351(6276):969–72.

- 日時: 9月26日(木)11:30 – 12:20
- 会場: D会場(天葉)
- 演者: Alex Rigort (Product Marketing Manager, Thermo Fisher Scientific)
- 演題: Cryo-tomography: a new imaging technique for cell biology to peer at the inner workings of cells
- 要旨:

Studying the molecular machinery of cells from atomic detail to the cellular context and beyond is a great challenge for cell biology. This luncheon will highlight how cryo-electron tomography allows to peer inside cells and see proteins in situ – in their unperturbed functional environments – at high resolution and in 3D.

The best possible structure preservation is guaranteed by sample vitrification, a freezing process so fast that it preserves structural integrity and functional interactions. Introducing innovative and user-friendly instrumentation makes this cutting-edge technology more applicable for cell biology. This lecture will introduce the suite of instruments that enable Thermo Fisher Scientific's cryo-electron tomography workflow as well as application examples from therecent literature.

お問い合わせ先:
日本エフイー・アイ株式会社
〒140-0002　東京都品川区東品川4-12-2
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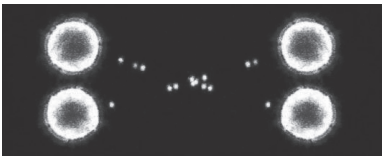
ThermoFisher SCIENTIFIC

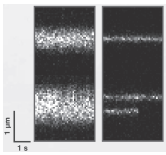
第 57 回　日本生物物理学会年会
日本カンタム・デザイン㈱　バイオフィジックスセミナー

LUMICKS
Capture Molecular Interactions

日時：9/26 (木) 11:30 - 12:20　　場所：F 会場 (4F, マーブル)
演者：Dr. Jordi Cabanas-Danés (Lumicks B.V.)

光ピンセット・蛍光顕微鏡システム　Lumicks C-Trap は、光ピンセットと一分子蛍光光学系を統合することにより、簡便にリアルタイム一分子計測ができるシステムです。すべての構成要素が一体成型の筐体に組み込まれることにより、ナノマニピュレーション・力学測定・蛍光測定いずれにおいても高い分解能・安定性・低ドリフト性を実現しています。光学系は共焦点蛍光顕微鏡だけでなく STED による超解像顕微鏡観察が可能です。





Confocal　STED

本発表では先進の光ピンセットシステムにより、どのような新たな発見ができるかをご紹介します。

Step into the Unresolved: Versatile Tools Towards Real-time Single-molecule Biology

Jordi Cabanas-Danés, Bas Groen and Andrea Candelli (Lumicks B.V.)
Sep. 26th (Thu)　11:30 - 12:20　　Place:　4F Room F (Marble)

Biological processes performed by proteins interacting with and processing DNA and RNA are key to cell metabolism and life. Detailed insights into these processes provide essential information for understanding the molecular basis of life and the pathological conditions that develop when such processes go awry.
The next scientific breakthrough consists in the actual, direct, real-time observations and measurements of the individual mechanisms involved, in order to validate and complete the current biological models.
Single-molecule technologies offer an exciting opportunity to meet these challenges and to study protein function and activity in real-time and at the single-molecule level.
Here, we present our efforts for further enabling discoveries in the field of biology and biophysics using both the combination of optical tweezers with single-molecule fluorescence microscopy (C-Trap).
We show the latest applications of these technologies that can enhance our understanding not only in the field of DNA/RNA-protein interactions but also in the fields of molecular motors, protein folding/unfolding, cell membranes and genome structure and organization.
These experiments show that the technological advances in hybrid single-molecule methods can be turned into an easy-to-use and stable instrument that has the ability to open up new venues in many research areas.

Quantum Design
JAPAN

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第57回日本生物物理学会年会　バイオフィジックスセミナー

SINCE 1889

微粒子・細胞の新しい分析法
磁気泳動と人工知能を使った画像解析

9 月 26 日（木）11 時 30 分〜 12 時 20 分　G 会場（ｱｲﾎﾞﾘｰ）

河野　誠

株式会社カワノラボ
大阪大学にて分析化学で博士号取得
電場や磁場を用いた粒子分析で起業
分析化学、界面科学
http://www.kawanoparticle.com

佐波　晶

大日本印刷株式会社
東京農工大学博士号取得（工学）。細胞・受精卵や電子顕微鏡の画像を対象に画像解析技術の開発に従事。
https://www.dnp.co.jp

磁気泳動による粒子 / 細胞評価（カワノラボ）

微粒子の磁気泳動から体積磁化率を 1 粒子ごとに求め、粒子表面への溶媒吸着体積、修飾体積、界面活性剤吸着体積から、粒子内部への包接体積など、粒子を構成する成分の体積を評価する新しい分析法。永久磁石で極所に高磁場 (3T) を作ることで、反磁性体でも泳動させることができ、細胞の種や活性状態も評価できる。また、結晶系の違いも評価できるなど、従来の電気泳動では得られない評価分析が可能であり、当日はこれらを紹介する。

人工知能を使った画像解析（大日本印刷）

近年、細胞や粒子などの評価は各種顕微鏡法を用いた”可視化”のニーズが高まっている。さらに、MS イメージング法や顕微ラマン法など分子情報得る装置の高度化、X 線 CT のような大きな構造体の断層写真など、画像データによる評価分析の可能性が広がっている。大日本印刷では、人工知能によって迅速かつ高精度な画像解析法を開発してきた。これまで、iPS 細胞の未分化・脱未分化領域をテクスチャ特徴量を用いて識別すの手法などを開発してきた。本発表では、最新の成果を実例をもって紹介する。

第57回日本生物物理学会年会
バイオフィジックスセミナー

BRUKER

● 日時： 2019年9月26日（木）　11：30～12：20

● 会場： H会場（アンバー）

高分解能質量分析による蛋白質の構造解析

演者　内山　進　先生

大阪大学大学院工学研究科生命先端工学専攻
自然科学研究機構生命創成探究センター(ExCELLS)

バイオ医薬品の主成分は医療用蛋白質であるが、蛋白質は、輸送や保管中、さらにはハンドリング中にストレスを受け、化学構造が変化することがある。特に、酸素による構成アミノ酸の酸化は、機能低下や副作用へとつながる可能性があることから、詳細な解析と適切な管理戦略が必要である。ブルカー製高分解能質量分析計maXis IIは、最高分解能のQ-ToF型質量分析装置であり、さらに同位体の強度比が正確に得られることから、質量値に基づいた構造解析を高い信頼性で実施出来る。
本セミナーではモノクローナル抗体の構造解析の例を用いながら、蛋白質研究における高分解能質量分析の有用性と可能性について紹介する。

maXis II

ブルカージャパン株式会社　ダルトニクス事業部

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【緊急告知】

年会２日目（9/25）懇親会の司会者はこの人

新進気鋭のヴァイオリニスト 松本尚子 さんです！

懇親会当日は 4F ホワイエにて生演奏も行います。

美しい音色をぜひライブでご堪能ください。

【演奏予定時刻】 11:10-11:30 (BP セミナー前、必聴！)

12:20-14:10 (時間調整有り、記念撮影も可)

日本生物物理学会主催
先端バイオイメージング支援プラットフォーム(ABiS) 共催

バイオフィジックスセミナー

イメージコンテスト

2019年 9月26日（木）11:30～12:30
E 会場
［シーガイアコンベンションセンター・クリスタル］

ABiSのご紹介

ABiSは科研費取得者を対象に、最先端のイメージング技術支援を進めています
その技術支援による成果や利用方法についてご説明いたします
演者：上野直人（基礎生物学研究所）
：真野昌二（基礎生物学研究所）

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文部科学省科学研究費助成事業・新学術領域研究・学術研究支援基盤形成
「先端バイオイメージング支援プラットフォーム（ABiS）」
（平成28年度 - 令和3年度）
研究支援代表者 狩野 方伸（自然科学研究機構 生理学研究所 / 東京大学）

ABiS
Advanced Bioimaging Support

本学会の連絡先は下記の通りです.

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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
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