NTHU-OU Virtual Symposium on Biological and Medical Sciences

January 19, 2022

大阪大学
OSAKA UNIVERSITY

2022 NTHU-OU Virtual Symposium on Biological and Medical Sciences
January 19, 2022

Zoom Link:
https://us02web.zoom.us/j/88544178723?pwd=T2xBMWY4dUY1dEdDdGtqdUZYSFdUdz09
Meeting ID: 885 4417 8723
Pass ward: 497080

Program in Japan Time (Taiwan Time)

10:00-10:05 (9:00-9:05) Opening Remarks: **Kenji Matsuno**, Professor, Department of Biological Sciences, OU

10:05-10:10 (9:05-9:10) Opening Remarks: **Ruey Ho Kao**, Professor and Dean, College of Life Science, NTHU

10:10-10:40 (9:10-9:40) **Tatsuo Kakimoto (Professor)**, OU
Patterning root tissues in *Arabidopsis thaliana*

10:40-11:00 (9:40-10:00) **Chang-Yi Chiu (PhD student)**, NTHU
Physiological role of the *Arabidopsis* CORNICHON HOMOLOG 5 (CNIH5) in nutrient homeostasis

11:00-11:30 (10:00-10:30) **Tzu-Kang Sang (Professor)**, NTHU
VCP ATPase safeguards genome and the nuclear structure: a sizable problem

Break

11:40-12:00 (10:40-11:00) **Kohei Watanebe (PhD student)**, OU
The optic lobe-pars intercerebralis axis is involved in circa'bi'dian rhythm of the large black chafer *Holotrichia parallela*

12:00-12:20 (11:00-11:30) **Christopher Llynard D. Ortiz (PhD student)**, NTHU
Ribosome in rolled state modulates-1 Programmed Ribosomal Frameshifting (-1 PRF)

12:20-12:40 (11:20-11:40) **Shunsuke Chuma (PhD studenet)**, OU
Elucidation of neural differentiation using intracellular temperature Imaging

Lunch 12:40-14:00 (11:40-13:00)
Drinks are exchanged between NTHU and OU

14:00-14:20 (13:00-13:20) **Min-Zong Liang (PhD student)**, NTHU

Investigate the role and the transcriptional regulation of PGAM5 during neuronal regeneration.

14:20-14:40 (13:20-13:40) **Zi Wang (Master student)**, OU

A dynamic molecular regulatory network to determine hippocampal cell fate during embryogenesis

14:40-15:00 (13:40-14:00) **Ming-Yang Li (PhD student)**, NTHU

The role of calcium uptake between ER and mitochondria in Srv2/CAP2 knockout SH-SY5Y cells

Break

15:10-15:30 (14:10-14:30) **Asuka Yamagichi (Master student)**, OU

Identification of protein motifs in MyosinIC and MyosinID responsible for left-right asymmetry of the hindgut in *Drosophila*

15:30-15:50 (14:30-14:50) **Grace Y. Liu (Undergraduate student)**, NTHU

A rapid and reversible tool to disassemble target microtubules

15:50-16:10 (14:50-15:10) **Hiroaki Nakajima (PhD student)**, OU

Redescription and phylogenetic analysis of *Octopus alatus* (Sasaki, 1920)

On Line Mixer 16:10-16:40 (15:10-15:40)

Snacks and drinks are exchanged between NTHU and OU

16:40-17:10 (15:40-16:10) **Takuro Nakagawa (Associate professor)**, OU

Heterochromatin suppresses gross chromosomal rearrangements at centromeres

17:10-17:40 (16:10-16:40) **Yu-Hsin Chiu (Assistant professor)**, NTHU

A non-canonical pathway for sympathetic activation of ATP-releasing Pannexin1 channels

17:40-18:00 (16:40-17:00) Ceremony of Awarding Best Presentations

18:00-18:05 (17:00-17:05) Closing Remarks: **Tatsuo Kakimoto**, Professor and Director, Department of Biological Sciences, OU

18:05-18:10 (17:05-17:10) Closing Remarks: **I-Ching Wang**, Associate Professor, The Institute of Biotechnology, NTHU

Patterning root tissues in *Arabidopsis thaliana*

Tatsuo Kakimoto

Department of Biological Sciences, Graduate School of Science, Osaka University
1-1 Machikaneyama, Toyonaka, Osaka 560-0043, Japan
e-mail: kakimoto@bio.sci.osaka-u.ac.jp

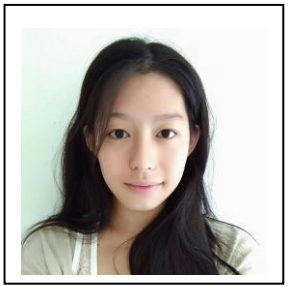
The vasculature system of plants consists of the water-transporting xylem and photoassimilates-transporting phloem. The phloem consists of the sieve elements (SEs), the apparatus for bulk flow, and the companion cells (CCs), which functions for loading and unloading. How proper pattern of these cell types are formed is yet to be clarified. We show that Dof-class transcription factors preferentially expressed in the phloem (phloem-*Dof*s) are necessary and sufficient for SE and CC differentiation. Phloem-Dofs not only induce genes necessary for SE and CC formation, but also induce genes for secretory signalling molecules, CLE25/26/45. CLEs are perceived by BAM–CIK receptor–coreceptor pairs and reduce phloem-Dof levels, forming a feedback loop. We propose that this Dof-CLE circuit creates the phloem pattern. I will also introduce our study on lateral root formation. Pericycle spontaneously forms auxin peaks, which in turn induces formative cell divisions. We identified transcription factors that govern the competency of pericycle cells to undergo auxin-induced lateral root initiation.

References and Footnotes

Zhang Y, Mitsuda N, Yoshizumi T, Horii Y, Oshima Y, Ohme-Takagi M, Matsui M, Kakimoto T (2021) Two types of bHLH transcription factor determine the competence of the pericycle for lateral root initiation. Nature Plants 7, 633-643.

Qian P, Song W, Yokoo T, Minobe A, Wang G, Ishida T, Sawa S, Chai J, Kakimoto T (2018) The CLE9/10 secretory peptide regulates stomatal and vascular development through distinct receptors. Nature plants 4,1071-1081

Physiological role of the *Arabidopsis* CNIH5 in nutrient homeostasis



Chang-Yi Chiu, Cheng-Da Tsai and Tzu-Yin Liu

Institute of Bioinformatics and Structural Biology, National Tsing Hua University, Hsinchu, Taiwan (R.O.C.)

e-mail: miya9630@gmail.com

Plants facilitate the uptake and translocation of Pi by increasing the expression of the plasma membrane (PM)-localized PHT1 Pi transporters. Although the ER export of several PHT1 family members is known to be assisted by PHF1, other accessory factors may be also involved in this process. The cornichon proteins are unique to eukaryotes and function as cargo receptors to help the package of several polytopic membrane proteins into the coat protein II (COPII) transport vesicles. Five *CORNICHON HOMOLOG (CNIH)* genes are identified in *Arabidopsis thaliana*. While *At*CNIH1 and *At*CNIH4 are responsible for subcellular targeting of the glutamate receptor-like channels implicated in Ca^{2+} homeostasis in pollen tube, the physiological functions of other CNIH members are unclear. In this study, we showed that *AtCNIH5* are specifically upregulated by Pi starvation. Public single cell RNA-seq and yeast split-ubiquitin interactome datasets indicated that *AtCNIH5*, *AtPHT1;1* and *AtPHF1* have similar tissue expression patterns and *At*CNIH1 interacts with *At*PHF1 and *At*PHT1;9. We thus speculate that *At*CNIH proteins, in particular *At*CNIH5, may serve as the cargo receptor for several PHT1s. Fluorescent tagging of *At*CNIH5 indicated its ER localization. Using the tripartite split-GFP assay in agro-infiltrated tobacco leaves and the yeast split-ubiquitin assay, we showed that *At*CNIH5 interacted with *At*PHT1;1. Measurement of Pi levels of the *cnih* single and double mutants implied *At*CNIHs may function redundantly to regulate Pi homeostasis. Our results will open a new avenue to study the post-translational regulation of PHT1 Pi transporters and delineate the physiological role of the plant cornichon family.

VCP ATPase safeguards genome and the nuclear structure: a sizable problem

Tzu-Kang Sang

Institute of Biotechnology, College of Life Science, National Tsing Hua University
101Kuang-Fu Rd. Sec. 2, Hsinchu, Taiwan 30013, Republic of China
e-mail: tksang@life.nthu.edu.tw

The change in the nuclear morphology serves as a prognostic hallmark in several malignant tumors and premature aging. However, insight into how a cell maintains nucleus morphology and whether nuclear structure alteration copes with physiological stresses remains poorly understood. We show the disruption of Drosophila homolog of human VCP ATPase causes nuclear size increase. VCP is essential for ubiquitin-dependent degradation or segregation of protein substrates. TER94 loss of function (LOF) leads to accumulations of ubiquitinated protein inside the expanding nucleus. Notably, the level of DNA damage indicators, γH2AV, and autophagy markers, ATG8a and p62, is also increased inside the expanding nuclei, linking defective DNA damage response and autophagy to nuclear size regulation. Genetic screens identify Mu2 (human MDC1 homolog) modify the enlarged nucleus defect caused by $TER94^{LOF}$, which is a TER94 substrate. We show that accumulated Mu2, in a p53A-dependent manner, increases nuclear size and autophagic markers. These genes function downstream of TER94 in this context, as knockdown of p53 and Mu2 suppresses the $TER94^{LOF}$-induced enlarged nuclei defect. Taken together, our results reveal that excessive DNA lesion, caused by retarded DNA repair due to the failure of removing Mu2 from chromatin by TER94, is a causal mechanism for aberrant nuclear size increase. We will discuss the interplays between autophagy and genomic integrity.

References and Footnotes

(1) Chang, Y.-C. et al. *Nature Communications* (12): 4258. (2021).

The optic lobe-pars intercerebralis axis is involved in circa'bi'dian rhythm of the large black chafer *Holotrichia parallela*

Kohei Watanabe

Department of Biological Sciences, Graduate School of Science, Osaka University
1-1 Machikaneyama, Toyonaka, Osaka 560-0043, Japan
e-mail: watanabe@bio.sci.osaka-u.ac.jp

The large black chafer *Holotrichia parallela* exhibits ~48-h circa'bi'dian rhythm. Although circabidian rhythm is suggested to involve the circadian clock, no physiological studies have been conducted to verify this involvement. Therefore, in order to investigate whether the brain regions required for circadian rhythm are also involved in circabidian rhythm, I examined the effects of optic lobe or pars intercerebralis removal on the circabidian rhythm. After removing both optic lobes, all beetles lost their circabidian rhythms (N=25), but all beetles exhibited circabidian rhythm after removing unilateral optic lobe (N=18). However, 22% of the latter group exhibited day switching. After removal of the pars intercerebralis, 26.3% beetles showed arrhythmic patterns (N=19). The number of paraldehyde fuchsin-stained pars intercerebralis cells in the arrhythmic group was significantly reduced compared to in the intact and sham-operated groups. The activity in the pars intercerebralis-removed beetles was significantly higher than that in the control groups. These results show that the optic lobe and at least part of the pars intercerebralis are necessary for circabidian rhythm, and bilateral optic lobes are necessary to maintain regularity of the two-day rhythm in *H. parallela*. This suggests that a neural circuit of circadian clock cells in the optic lobe to pars lateralis might be evolutionally conserved and used also for the generation of circabidian rhythm.

Ribosome in Rolled state modulates -1 Programmed Ribosomal Frameshifting (-1 PRF)

Christopher Llynard Ortiz[1,2,3†], **Yi-Lan Chen**[5], **Irene Deli**[5], **Jin-Der Wen**[4], **Ming-Daw Tsai**[5], **Lee-Wei Yang**[2,6]

[1]Department of Chemistry, National Tsing Hua University, Hsinchu 30013, Taiwan

[2]Institute of Bioinformatics and Structural Biology, National Tsing Hua University, Hsinchu 30013, Taiwan

[3]Chemical Biology and Molecular Biophysics Program, Taiwan International Graduate Program, Institute of Biological Chemistry, Academia Sinica, Taipei 115, Taiwan

[4]Institute of Molecular and Cellular Biology, National Taiwan University, Taipei 10617, Taiwan

[5]Institute of Biological Chemistry, Academia Sinica, Taipei 115, Taiwan

[6]Physics Division, National Center for Theoretical Sciences, National Taiwan University, Taipei 10617, Taiwan

† e-mail: cldortiz@gapp.nthu.edu.tw

Ribosomes, the biological translational machinery, decodes the information from the mRNA into proteins. This machinery is typically hijacked by pathogens like viruses to produce viral proteins necessary for their proliferation and survival. One of the mechanisms they have developed is known as the programmed ribosomal frameshifting, specifically -1 programmed ribosomal frameshifting (-1 PRF). This mechanism allows the production of a different set of proteins from a single mRNA. Although slippery sequence and structured mRNA (i.e., pseudoknot and hairpin) are known to be the key elements in driving this mechanism, their effect on the ribosome's conformational dynamics remains elusive.

To reveal the dynamics during -1 PRF, we have previously published a paper that showed a model using *T. thermophilus* ribosome and theoretical techniques (i.e., linear response theory and resolution-exchange simulations) to reconcile existing information about -1 PRF from structural, single-molecule, and mutagenesis data. We discover that the presence of a pseudoknot (PK) in the mRNA entrance tunnel generates resistant forces that makes the 30S adopt a new conformational state, through subunit rolling. This motion is orthogonal to the 30S body rotation and is different from the hyper-rotation motion (>8° 30S body rotation) that was proposed earlier when the ribosome encountered a structured mRNA. Further results suggested that in the presence of a PK with ~50% frameshifting efficiency, the 30S subunit rolling angle was twice more as compared to the PK having 0% frameshifting efficiency. To further prove this concept, we use cryo-EM to capture the *E. coli* ribosome structure upon encountering a human telomerase pseudoknot (DU177) and its mutant, U3C. Flexible fitting was utilized to elucidate the all-atom structure of each ribosomal state captured by cryo-EM. Interestingly, we found that one of the ribosome states with DU177 PK has a 30S subunit rolling angle of ~4.67° which is twice more as compared to when the ribosome encountered U3C PK (~1.96°). Additionally, our cryo-EM maps revealed at least 3 states of pseudoknot which are in before, during and after its unfolding. So far, these PK maps are the clearest experimentally resolved ones which could give us an insight on the pseudoknot unfolding mechanism involving corresponding conformational changes in the ribosome. Indeed, our cryo-EM data suggests that the ribosome undergoes 30S subunit rolling to modulate the -1 PRF as what we predicted two years ago.

References and Footnotes

(1) Chang, Kai-Chun, et al. *Bioinformatics* 35.6: 945-952 (2019).

(2)Yang, Lee-Wei, et al. *Biophysical journal* 107.6: 1415-1425 (2014).

Elucidation of Neural Differentiation Using Intracellular Temperature Imaging

Shunsuke Chuma

Department of Biological Sciences, Graduate School of Science, Osaka University
3-2 Yamadaoka, Suita, Osaka 565-0871, Japan
e-mail: schuma@protein.osaka-u.ac.jp

Neural differentiation is an essential process for brain development. Neural stem cells differentiate into neurons with neurite outgrowth, regulated by extracellular factors (e.g. cytokine) and/or intracellular factors (e.g. DNA methylation or histone modification). Recent studies have shown that thermal stimulation by increasing the culture temperature influences neural differentiation[1]. However, changes in intracellular temperature during neural differentiation and the detailed neural differentiation mechanisms involved in intracellular temperature remain elusive.

Here, we investigated the changes in intracellular temperature during neural differentiation of PC12 cells and the influence of intracellular temperature on neural differentiation. Using a fluorescence polymeric thermometer which shows the elongation of fluorescence lifetime as temperature rises[2] and fluorescence lifetime imaging microscopy, we found that the intracellular temperature increased by approximately 1 °C during differentiation in a transcription and translation dependent manner. Additionally, local heating of the nucleus with an infrared laser (ΔT = +2~5°C) during induction of neural differentiation promoted neurite outgrowth. Therefore, we propose that thermogenesis in the nucleus during differentiation contributes to neurite outgrowth.

References and Footnotes

1) Kudo, T. *et al.*, *PLOS ONE* 10, e0124024, (2015)
2) Okabe, K. *et al.*, *Nature Communications* 3, 705-713 (2012).

Investigate the role and the regulation of PGAM5 during neuronal regeneration

Min-Zong Liang

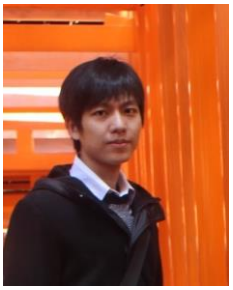
Institute of Molecular Medicine, National Tsing Hua University,
No.101, Sec. 2, Guangfu Rd., East Dist., Hsinchu City 300, Taiwan
e-mail: liang4646@gmail.com

Traumatic brain injury (TBI) is a public health problem with high mortality and morbidity. There are about 70 million individuals worldwide suffering TBI every year. However, there is currently no effective treatment for TBI due to limited regenerative potential of damaged brain neurons. We have showed that mitochondrial transplantation and recombinant WNT3A protein promote neuronal regeneration upon injury[1,2]. However, it is unclear whether the promoted neuronal regeneration induced by mitochondria therapy and WNT3A treatment are related. It has been reported that phosphoglycerate mutase 5 (PGAM5) released from damaged mitochondria can activate WNT signaling and promote mitochondria biogenesis. We hypothesized that PGAM5 might be the modulator in the interplay between WNT signaling and mitochondria biogenesis. We found PGAM5 was upregulated and released to cytosol in neurons after injury. We further investigated whether PGAM5 modulates mitochondrial biogenesis and neuronal regeneration upon injury. Since PGAM5 was upregulated during neuronal regeneration, we next investigated the transcriptional regulation of PGAM5. We predicted potential enhancer elements of *Pgam5* and assessed eRNA expression levels to identified activated enhancer regions during neuronal regeneration. Together, my study is to investigate whether PGAM5 modulates mitochondrial biogenesis and neuronal regeneration and the transcriptional regulation of PGAM5 during neuronal regeneration.

References and Footnotes

(1) Chien et al. *Biochim Biophys Acta Mol Basis Dis.*1864(9 Pt B):3001-3012 (2018)
(2) Chang et al. *Int J Mol Sci.* 21(4):1463 (2020)

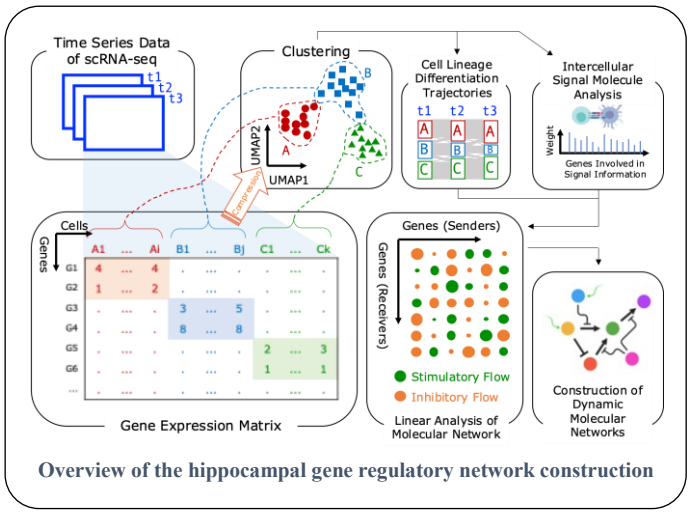
A Dynamic Molecular Regulatory Network to Determine Hippocampal Cell Fate during Embryogenesis



Zi Wang

Laboratory for Cell Systems, Institute for Protein Research, Osaka University
3-2 Yamadaoka, Suita-shi, Osaka 565-0871, Japan
e-mail: wang.zi@protein.osaka-u.ac.jp

Finding the mechanisms that determine cell fate in the hippocampus during embryonic development will help us understand memory formation. Previous studies identified key regulatory molecules at hippocampal developmental stages [2]. However, the full picture of molecular interactions and regulatory networks relevant to hippocampal development remains elusive. In this study, we aim to establish a systematic analytical method to integrate time-series scRNA-seq data into interpretable gene regulatory networks (GRNs), revealing intermolecular regulatory mechanisms during embryonic hippocampal cell differentiation.



Overview of the hippocampal gene regulatory network construction

We performed unsupervised clustering [1] of time-series scRNA-seq data from developing hippocampus [4] and identified 21 cellular clusters, then quantified differentiation trajectories by k-nearest neighbor method, then found 4 intercellular signaling groups by CellChatDB [3], and finally made 12 GRNs from gene expression patterns in pre- and post-temporal causal states.

By comparing the GRNs constructed for each cell lineage, we propose specific gene regulatory patterns may dominate points of divergence that determine differentiation between neuronal and glial cells. Furthermore, based on the large differences in patterns between adjacent developmental stages of the same lineage, we suggest that key regulatory factors may be present at early developmental stages.

References and Footnotes

[1] Butler A. *et al.*, *Nat Biotechnol*, 36:411-420, 2018.
[2] Kempermann G. *et al.*, *Trends Neurosci*, 27:447-452, 2004.
[3] Jin, S. *et al.*, Nat Commun, 12:1088, 2021.
[4] Zhong, S. *et al.*, *Nature*, 577:531-536, 2020.

Mechanistic study of the role of calcium signaling between ER and mitochondria by Srv2/CAP2 knockout mammalian cells

Ming-Yang Li[1], Chuang-Rung Chang[1,2]

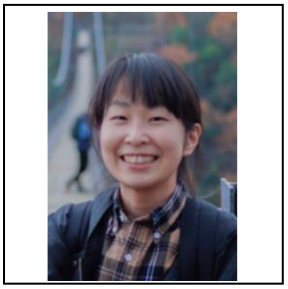
[1]Institute of Biotechnology, National Tsing-Hua University, Hsinchu, Taiwan
[2]Department of Medical Science, National Tsing-Hua University, Hsinchu, Taiwan
e-mail: crchang@life.nthu.edu.tw

Mitochondria are critical organelles that regulate cellular metabolism and produce energy currency of the cell through respiration. They are highly dynamic cellular organelles. Changes in the number and size of mitochondria in a cell are controlled by two opposing forces: fission and fusion. Fusion factor is MFN1, MFN2 and OPA1. Fission factor is Drp1. Mitochondria are connected with many cellular organelles, the best-characterized membrane contact sites bridge the endoplasmic reticulum (ER) and mitochondria. ER and mitochondria are tightly associated with very dynamic platforms termed mitochondria-associated membranes (MAMs). MAMs are involved in the transport of calcium from the ER to mitochondria. VDACs are located at the outer mitochondrial membrane (OMM). Mitochondrial calcium uniporter (MCU) accumulates of calcium from intermembrane space to the mitochondrial matrix. We previously characterized Srv2/CAP2, which is an actin binding protein involved in the regulation of mitochondria dynamics. The current evidence proves that Srv2/CAP2 interacts with the mitochondrial fission protein DRP1 and functions as a pro-fission factor. We hypothesized that Srv2/CAP2 mediated mitochondria dynamics may affect calcium uptake. To clarify the roles of Srv2/CAP2 in calcium uptake in the regulation of mitochondrial dynamics and ER/mitochondria contact, we applied CEPIA, which emits green fluorescence while binding to calcium in mitochondria and ER. We have found that more ER and mitochondria calcium contact sites in Srv2/CAP2 knockout cells. We expect to elucidate calcium flux under stress conditions and clarify the role of organellar calcium in the regulation of mitochondria after successful execute the proposed experiments.

References and Footnotes

(1) Chen, Y. C., et al. *Iscience* 11: 305-317(2019).

Identification of protein motifs in MyosinIC and MyosinID responsible for left-right asymmetry of the hindgut in *Drosophila*



Asuka Yamaguchi

Department of Biological Sciences, Graduate School of Science, Osaka University
1-1 Machikaneyama, Toyonaka, Osaka 560-0043, Japan
e-mail: ayamaguchi@cm.bio.sci.osaka-u.ac.jp

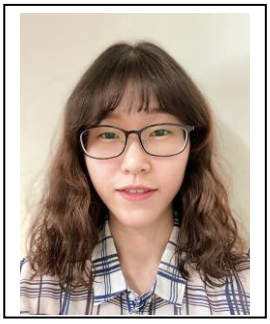
Many animals have directional left-right (LR) asymmetric structures in their bodies. However, molecular mechanisms of the LR asymmetric development in invertebrates are still unknown. *Drosophila* shows LR asymmetry in the embryonic hindgut [(1)]. The hindgut is formed asymmetrically due to the chirality of epithelial cells in the hindgut, which we call cell chirality [(2)]. *Drosophila* MyosinIC (MyoIC) and MyosinID (MyoID), which belong to the conserved Myosin I family, have opposing activities for directing the LR asymmetry of the hindgut and for defining the enantiomorphic states of cell chirality: MyoIC and MyoID have sinistral and dextral activities, respectively.

Given that MyoIC and MyoID have crucial roles dictating cell chirality, we speculated that some structural motifs in these two proteins may act as sources for defining two chirality states or play essential roles in cell chirality formation. Previous research revealed that their head domains dictate the states of chirality [(3)]. Thus, we studied the roles of four loop regions, cm loop, loop 2, loop 3, and loop 4, which are part of the head domains where they serve as interfaces with F-actin. For this purpose, we swapped each loop region between MyoIC and MyoID and analyzed its effect on LR asymmetric development of the hindgut in the resulting chimeras. The results showed, that there were different activities depending on the loop region, whereas swapping of corresponding loops did not reverse their intrinsic function in dictating the LR asymmetric formation of the hindgut. Suggesting that a single loop region is not enough to trigger chirality. Here, we plan to discuss the experiments with mutants where multiple loops have been swapped.

References and Footnotes

(1) Hozumi, S. *et al*. *Nature* 440, 798-802 (2006).
(3) Taniguchi, K. *et al*. *Science* 333, 339-341 (2011).
(4) Hozumi, S. *et al*. *Developmental Dynamics* 237, 3528-3537 (2008).

A rapid and reversible tool to disassemble target microtubules



Grace Y. Liu

Interdisciplinary Program of Life Science, National Tsing Hua University
101, Section 2, Kuang-Fu Road, Hsinchu 300044, Taiwan R.O.C.
e-mail: gracey.liu215@gmail.com

Microtubules (MTs) are components of the evolutionarily conserved cytoskeleton, which tightly regulates various cellular activities[1-2]. Our understanding of MTs is largely based on MT-targeting agents[3], which, however, are insufficient to dissect the dynamic mechanisms of specific MT populations due to their slow effects on the entire pool of MTs. To address this limitation, we have used chemogenetics and optogenetics to disassemble specific MT subtypes including tyrosinated MTs, primary cilia, mitotic spindles, and intercellular bridges, by rapidly recruiting engineered MT-cleaving enzymes onto target MTs in a reversible manner. Acute MT disassembly swiftly halted vesicular trafficking and lysosomal dynamics. It also immediately triggered Golgi and ER reorganization and slowed the fusion/fission of mitochondria without affecting mitochondrial membrane potential. Cell rigidity was increased after MT disruption owing to increased contractile stress fibers. MT disruption prevented cell division but did not cause cell death during interphase. These tools enable to uncover new insights of how MTs precisely regulate cellular architectures and functions.

References and Footnotes

(1) Cleary, J.M., and Hancock, W.O. *Current Biology* 31, R560–R573 (2021).
(2) Janke, C., and Magiera, M.M. *Nature Reviews Molecular Cell Biology* (2020).
(3) Steinmetz, M.O., and Prota, A.E. *Trends in Cell Biology* 28, 776–792 (2018).

Redescription and phylogenetic analysis of *Octopus alatus* (Sasaki, 1920)

Hiroaki Nakajima

Department of Biological Sciences, Graduate School of Science, Osaka University
1-1 Machikaneyama, Toyonaka, Osaka 560-0043, Japan
e-mail: hnakajima@bio.sci.osaka-u.ac.jp

Octopuses are one of the important resources in Japanese fisheries. The current taxonomy of octopus is confusing and many octopus species remain to be described. Small to medium-sized octopuses caught as bycatch in bottom trawling are rarely distributed in the market, and are treated simply as "octopus" or "small octopus". In this study, we report new taxonomic findings on octopuses caught as bycatch in the offshore trawl fishery at a depth of 100-200 m in Saga fishing port, Kuroshio, Kochi, Shikoku, Japan. Octopus species by the trawl fishery are followings: *Amphioctopus areolatus* (de Haan [in Férussac & d'Orbigny], 1839-1841), *Callistoctopus minor* (Sasaki, 1920), and *Octopus alatus*. *Octopus alatus* is similar to *C. minor*, but it is distinguishable by having fresh body coloration, supra-ocular cirrus, basal chromatophore, w-shaped funnel organ, conical hectocotylus, and 70—80 suckers on the hectocotylized arm. *Octopus alatus* was described by Sasaki (1920) based on only two males collected in the Bungo Channel, Shikoku, Japan, without female specimens. In this study, we collected adequate specimens including females to describe the characteristics of this species. Further, we inferred the COX 1 gene phylogeny of Octopoda to study the phylogenetic affinity of *Octopus alatus*.

References and Footnotes

(1) Sasaki, M. *Proceedings of the United States National Museum* 57, 163-203 (1920).

Heterochromatin suppresses gross chromosomal rearrangements at centromeres

Takuro Nakagawa

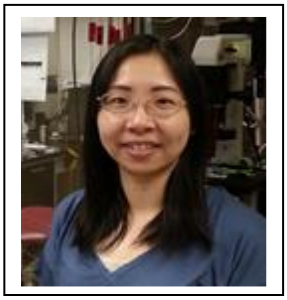
Department of Biological Sciences, Graduate School of Science, Osaka University
1-1 Machikaneyama, Toyonaka, Osaka 560-0043, Japan
e-mail: takuro4@bio.sci.osaka-u.ac.jp

Heterochromatin characterized by histone H3 lysine 9 (H3K9) methylation is formed at centromeres that consist of repetitive sequences and represses transcription of centromeric noncoding RNAs. In mice, loss of the H3K9 methyltransferase causes chromosome instability and cancer predisposition[1]. However, it remains unclear how heterochromatin maintain chromosome stability. Using fission yeast, we found that heterochromatin suppresses gross chromosomal rearrangements (GCRs) at centromeres by repressing transcription[2]. Loss of Clr4, the H3K9 methyltransferase in fission yeast, increased spontaneous rates of GCRs by around one hundred-fold. Changing the H3K9 residue to alanine (H3K9A) or arginine (H3K9R) also increased GCR rates, demonstrating that Clr4 suppresses GCRs through H3K9 methylation. Physical analysis of chromosomes revealed that the *clr4* deletion increases the formation of isochromosomes whose arms mirror each other. Breakpoints were found in centromere inverted repeats, showing that recombination between the centromere repeats produces isochromosomes. Interestingly, mutation in the catalytic subunit of RNA polymerase II, Rpb1, or the transcription factor, Tfs1/TFIIS, strongly reduced GCR rates in c*lr4*Δ cells, showing that heterochromatin suppresses centromeric GCRs via the transcription regulation. We would like to share our recent data and discuss how transcription causes GCRs at centromeres.

References and Footnotes

(1) Peters, A.H.. et al. *Cell* 107, 323-337 (2001).
(5) Okita, A.T. et al. *Commun Biol* 2, 17 (2019).

A non-canonical pathway for sympathetic activation of ATP-releasing Pannexin1 channels



Yu-Hsin Chiu

Institute of Biotechnology, Departments of Medical Science and Life Science, National Tsing Hua University
101, Sec. 2, Kuang Fu Rd., Hsinchu, Taiwan 300044
e-mail: yhchiu@life.nthu.edu.tw

Pannexin 1 (Panx1) constitutes cell-surface ion channels that can be found in diverse vertebrate cells of immune, cardiovascular, or neuronal systems. Unlike classical ion channels that can only permeate inorganic ions, activated Panx1 channels can also allow the release of intracellular metabolites (e.g., ATP or spermidine) to mediate cell-to-cell communications. For example, ATP released via Panx1 channels can engage purinergic signaling in many physiological conditions, and thus aberrant Panx1 activities have been implicated in multiple diseased states, such as neuropathic pain, seizure, hypertension, and inflammation. However, how Panx1 channels are activated in varying patho-physiological contexts remains unclear. We previously reported two distinct activation modalities, irreversible activation mediated by caspase 3/7 cleavage for apoptotic corpse removal and reversible activation by α1 adrenoceptor (α1-AR) signaling for sympathetic vasoconstriction. In this presentation, I focus on describing a non-canonical pathway of α1-AR, which involves Gq, RhoGTPase and mDia, as well as HDAC6-mediated lysine deacetylation of Panx1 channels. These findings uncovered a new activity switch of ion channels by acetylation-deacetylation, and presented potential targets for developing the pathway-specific therapeutics of Panx1-associated diseases.

References and Footnotes

(1) Chiu, YH. et al. *Nature Communications* 12(1):4482 (2021).
(6) Chiu, YH. et al. *Journal General Physiology* 150(1):19-39 (2018).