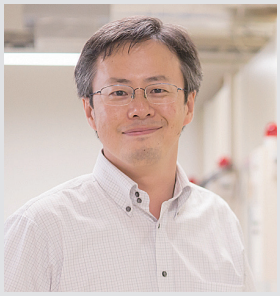
RESEARCH ACTIVITIES
Research Center of Integrative Molecular Systems

The mission of CIMoS is to analyze molecular systems in nature to find the logic behind the sharing and control of information between the different spatiotemporal hierarchies, with the ultimate goal of creating novel molecular systems on the basis of these findings.

Biological Rhythm and Dynamics through Chemistry

Research Center of Integrative Molecular Systems
Division of Trans-Hierarchical Molecular Systems



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Keywords Biological Rhythm, Circadian Clock, Cyanobacteria

Living organisms on Earth evolved over time to adapt to daily environmental alterations, and eventually acquired endogenous time-measuring (biological clock) systems. Various daily activities that we perform subconsciously are controlled by the biological clock systems sharing three characteristics. First, the autonomic rhythm repeats with an approximately 24-hour (circadian) cycle (self-sustainment). Second, the period is unaffected by temperature (temperature compensation). Third, the phase of the clock is synchronized with that of the outer world in response to external stimuli (synchronization). We seek to explain these three characteristics, and consider the biological clock system of cyanobacteria to be an ideal experimental model.

The major reason that cyanobacteria are considered to be the ideal experimental model is that the core oscillator that possesses the three characteristics of the clock can be easily reconstructed within a test tube. When mixing the three clock proteins KaiA, KaiB, and KaiC with ATP, the structure and enzyme activity of KaiC change rhythmically during a circadian cycle. Taking advantage of this test tube experiment, we used an approach combining biology, chemistry, and physics to elucidate the means by which the clock system extends from the cellular to atomic levels.

Among the three Kai proteins, KaiC is the core protein of the oscillator. In the presence of KaiA and KaiB, KaiC revelas the rhythm of autophosphorylation and dephosphorylation; however, the cycle of this rhythm depends on the ATPase activity of KaiC independent of KaiA or KaiB. For example, when the ATPase activity of KaiC doubles as a result of amino acid mutations, the frequencies of both the *in vitro* oscillator and the intracellular rhythm also double (the cycle period is reduced to half). This mysterious characteristic is called a transmural hierarchy, in which the cycle (frequency) and even the temperature compensation both *in vitro* and *in vivo* are greatly affected (controlled) by the function and structure of KaiC.

How are the circadian activities and temperature compensation features encoded in KaiC and then decoded from it to propagate rhythms at the cellular level? We are committed to better understanding biological clocks and other dynamic systems through the chemistry of circadian ***rhythm***, ***structure***, and evolutionary ***diversity***.

Selected Publications

- A. Mukaiyama, Y. Furuike, K. Ito-Miwa, Y. Onoue, K. Horiuchi, K. Kondo, E. Yamashita and S. Akiyama, *Nat. Commun.* **16**, 4541 (2025).
- Y. Furuike, Y. Onoue, S. Saito, T. Mori and S. Akiyama, *PNAS Nexus* **4**, pgaf136 (2025).
- Y. Furuike, A. Mukaiyama, S. Koda, D. Simon, D. Ouyang, K. Ito-Miwa, S. Saito, E. Yamashita, T. Nishiwaki, K. Terauchi, T. Kondo and S. Akiyama, *Proc. Natl. Acad. Sci. U. S. A.* **119**, e2119627119 (2022).
- Y. Furuike, A. Mukaiyama, D. Ouyang, K. Ito-Miwa, D. Simon, E. Yamashita, T. Kondo and S. Akiyama, *Sci. Adv.* **8**, eabm8990 (2022).
- J. Abe, T. B. Hiyama, A. Mukaiyama, S. Son, T. Mori, S. Saito, M. Osako, J. Wolanin, E. Yamashita, T. Kondo and S. Akiyama, *Science* **349**, 312–316 (2015).
- Y. Murayama, A. Mukaiyama, K. Imai, Y. Onoue, A. Tsunoda, A. Nohara, T. Ishida, Y. Maéda, T. Kondo and S. Akiyama, *EMBO J.* **30**, 68–78 (2011).

1. *Structure*: Reasons for Seeking Structure and Dynamics of Circadian Clock Components in Cyanobacteria[1–4)]

A great deal of effort has been devoted to characterizing structural changes in the clock proteins along the circadian reaction coordinate. However, little is known about the mechanism driving the circadian cycle, even for the simple cyanobacterial protein KaiC that has ATPase and dual phosphorylation sites in its N-terminal C1 and C-terminal C2 domains, respectively. Nearly all KaiC structures reported to date share a nearly identical structure, and they do not appear to be suggestive enough to explain the determinants of circadian period length and its temperature compensation. We are studying the structural and dynamical origins in KaiC using high-resolution x-ray crystallography,[1–4)] real-time fluorescence detection,[5)] and quasielastic neutron scattering.[6)]

2. *Rhythm*: Cross-Scale Analysis of Cyanobacterial Circadian Clock System[6–8)]

KaiC ATPase is of particular interest here, as it finely correlates to the frequencies of *in vivo* as well as *in vitro* oscillations and also it is temperature compensated. This unique property has inspired us to develop an ATPase-based screening[7)] for KaiC clock mutants giving short, long, and/or temperature-dependent periods.[8)] A developed HPLC system with a 4-channel temperature controller has reduced approximately 80% of time costs for the overall screening process (Figure 1). Using the developed device, we are screening a number of temperature-dependent mutants of KaiC.[6,7)]

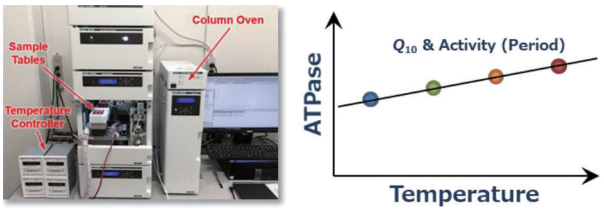

Figure 1. Development of a quick ATPase assay system.

3. Beyond Evolutionary *Diversity*[9)]

In the presence of KaiA and KaiB, the ATPase activity of KaiC oscillates on a 24-hour cycle. KaiC is not capable of maintaining a stable rhythm on its own, but its activity was observed to fluctuate with reduced amplitude over time (Figure 2A). We have identified a signal component that is similar to damped oscillation, and propose that it encodes the specific frequency, equivalent to a 24-hour cycle.

The habitats of cyanobacteria are diverse, so the space of their sequence is immense. Furthermore, some KaiA and KaiB genes are missing in several strains of cyanobacteria. This is understandable to some extent if KaiC possesses the specific frequency. Given this assumption, *what specific frequencies are possessed by KaiC homologues in other species and ancestral cyanobacteria?* (Figure 2B)

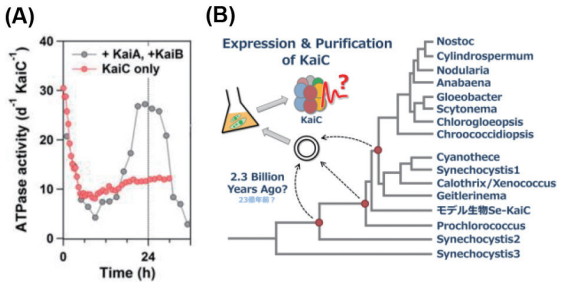

Figure 2. Damped oscillation of KaiC ATPase activity (**A**) and evolutionary diversity of cyanobacteria (**B**).

To address these questions, we restored the amino acid sequences of ancestral Kai proteins (Figure 2B) and studied their function (oscillation) and structures to determine the evolutionary origin of the self-sustained Kai-protein oscillators.[9)] Our results clearly demonstrate that the oldest Kai-protein oscillator emerges in the most recent common ancestor (MRCA) of cyanobacteria at approximately 2.2 Ga ago and is able to synchronize with temperature cycles of 18 h; this is shorter than the current rotation period of the Earth.

4. Bio-SAXS Activity in IMS[10)]

We have supported SAXS users so that they can complete experiments smoothly and publish their results.[10)]

References

1) Y. Furuike, A. Mukaiyama, D. Ouyang, K. Ito-Miwa, D. Simon, E. Yamashita, T. Kondo and S. Akiyama, *Sci. Adv.* **8**, eabm8990 (2022).
2) Y. Furuike, A. Mukaiyama, S. Koda, D. Simon, D. Ouyang, K. Ito-Miwa, S. Saito, E. Yamashita, T. Nishiwaki-Ohkawa, K. Terauchi, T. Kondo and S. Akiyama, *Proc. Natl. Acad. Sci. U. S. A.* **119**, e2119627119 (2022).
3) Y. Furuike, E. Yamashita and S. Akiyama, *Biophys. Physicobiol.* **21**, e210001 (2024).
4) Y. Furuike, Y. Onoue, S. Saito, T. Mori and S. Akiyama, *PNAS Nexus* **4**, pgaf136 (2025).
5) A. Mukaiyama, Y. Furuike, E. Yamashita and S. Akiyama, *Biochem. J.* **479**, 1505–1515 (2022).
6) Y. Furuike, D. Ouyang, T. Tominaga, T. Matsuo, A. Mukaiyama, Y. Kawakita, S. Fujiwara and S. Akiyama, *Commun. Phys.* **5**, 75 (2022).
7) D. Ouyang, Y. Furuike, A. Mukaiyama, K. Ito-Miwa, T. Kondo and S. Akiyama, *Int. J. Mol. Sci.* **20**, 2789–2800 (2019)
8) D. Simon, A. Mukaiyama, Y. Furuike and S. Akiyama, *Biophys. Physicobiol.* **19**, e190008 (2022).
9) A. Mukaiyama, Y. Furuike, K. Ito-Miwa, Y. Onoue, K. Horiuchi, E. Yamashita and S. Akiyama, *Nat. Commun.* **16**, 4541 (2025).
10) T. Inobe, R. Sakaguchi, T. Obita, A. Mukaiyama, S. Koike, T. Yokoyama, M. Mizuguchi and S. Akiyama, *FEBS Lett.* **598**, 2292–2305 (2024).

* IMS International Internship Program

Elucidation of Function, Structure, and Dynamics of Condensed-Phase Molecular Systems by Advanced Ultrafast Laser Spectroscopy

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Keywords Ultrafast Spectroscopy, Nonlinear Spectroscopy, Chemical Reaction Dynamics

We develop and apply advanced ultrafast laser spectroscopy based on state-of-the-art optical technology to study the chemical reaction dynamics of the condensed-phase molecules. In particular, we focus on exploiting unique methodologies based on few-cycle ultrashort pulses (*e.g.*, time-domain impulsive vibrational spectroscopy and multidimensional spectroscopy) and tracking molecular dynamics from electronic and structural viewpoints throughout the chemical reaction with exquisite temporal resolution. We also develop a novel methodology and light source to probe ultrafast dynamics of single molecules in the condensed phase at room temperature, with the aim to understand chemical reaction dynamics at the single-molecule level. Our particular interest rests on elucidating sophisticated molecular mechanisms that underlie the reactions of functional molecular systems such as proteins, molecular assemblies, and metal complexes. On the basis of new insights that can be gained from our advanced spectroscopic approaches, we aim to establish a new avenue for the study of chemical reaction dynamics.

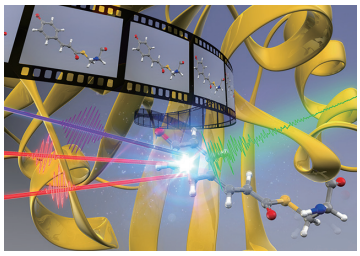

Figure 1. Schematic of the ultrafast nonlinear spectroscopy of complex molecules with few-cycle ultrashort pulses.

Selected Publications

- Y. Yoneda, T. Konishi, K. Suga, S. Saito and H. Kuramochi, "Excited-State Aromatization Drives Nonequilibrium Planarization Dynamics," *J. Am. Chem. Soc.* **147**, 12051 (2025).
- H. Kuramochi, T. Tsutsumi, K. Saita, Z. Wei, M. Osawa, P. Kumar, L. Liu, S. Takeuchi, T. Taketsugu and T. Tahara, "Ultrafast Raman Observation of the Perpendicular Intermediate Phantom State of Stilbene Photoisomerization," *Nat. Chem.* **16**, 22 (2024).
- Y. Yoneda and H. Kuramochi, "Room-Temperature Solution Fluorescence Excitation Correlation Spectroscopy," *J. Phys. Chem. Lett.* **15**, 8533 (2024).
- Y. Yoneda, and H. Kuramochi, "Rapid-Scan Resonant Two-Dimensional Impulsive Stimulated Raman Spectroscopy of Excited States," *J. Phys. Chem. A* **127**, 5276–5286 (2023).
- H. Kuramochi and T. Tahara, "Tracking Ultrafast Structural Dynamics by Time-Domain Raman Spectroscopy," *J. Am. Chem. Soc.* **143**, 9699–9717 (2021).
- H. Kuramochi, S. Takeuchi, K. Yonezawa, H. Kamikubo, M. Kataoka and T. Tahara, "Probing the Early Stages of Photoreception in Photoactive Yellow Protein with Ultrafast Time-Domain Raman Spectroscopy," *Nat. Chem.* **9**, 660–666 (2017).

1. Excited-State Aromatization Drives Non-Equilibrium Planarization Dynamics[1)]

Excited-state aromaticity is one of the most widely applied concepts in chemistry, often used as a rational guideline for predicting conformational changes in cyclic π-conjugated systems induced by photoexcitation. Yet, the details of the relationship between the corresponding photoinduced electronic and structural dynamics have remained unclear. In this work, we applied femtosecond transient absorption and time-resolved time-domain Raman spectroscopies to track a non-equilibrium planarization dynamics of cyclooctatetraene (COT) derivative associated with the excited-state aromaticity. In the femtosecond time-resolved Raman data, the bent-to-planar structural change was clearly captured as a continuous peak shift of the marker band, which was unambiguously identified with ^{13}C-labeling. Our findings show that the planarization occurs after a significant change in the electronic structure, suggesting that the system first becomes aromatic, followed by a conformational change. This work provides a unique framework for understanding the excited-state aromaticity from a dynamical aspect.

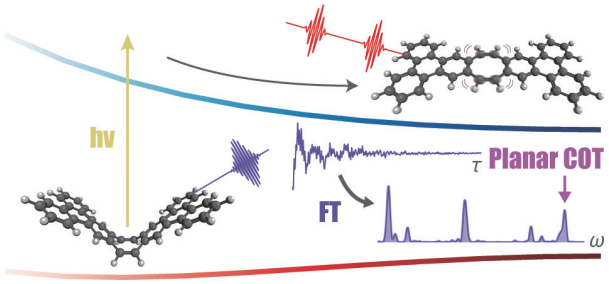

Figure 2. Schematic of time-domain Raman observation of the structural dynamics upon the onset of excited-state aromaticity.

2. Dynamic Excited-State Localization Induced by Jahn-Teller Distortion Observed by Coherent Vibrational Spectroscopy[2)]

Molecular symmetry is a central design element in functional materials, yet its dynamic modulation in the excited state and its consequences for optoelectronic properties remain largely unexplored, particularly in main-group p-block element complexes. We address this knowledge gap by investigating unique Al(III) dinuclear triple-helical complexes that combine high symmetry with twisted π-conjugated systems and achieve exceptional optical properties of unusually large Stokes shifts and high photoluminescence quantum yields. Using transient absorption spectroscopy with a 10 fs pump pulse, we detected coherent vibrational oscillations overlapped with transient absorption/stimulated emission signals. Analysis of the dephasing times of oscillatory signals revealed photoexcitation-triggered Jahn-Teller distortions in these high-symmetry p-block complexes, evidenced by a specifically short dephasing time constant of 410 fs associated with intraligand twisting vibrations. Our findings demonstrate that excited-state symmetry breaking, strongly coupled with intraligand twisting vibrations, is crucial in determining the remarkable photofunctional properties of large Stokes shifts and high photoluminescence quantum yields. This work elucidates the fundamental mechanisms underlying the performance of these Al(III) complexes and provides a conceptual framework for designing next-generation photofunctional materials by harnessing dynamic symmetry changes.

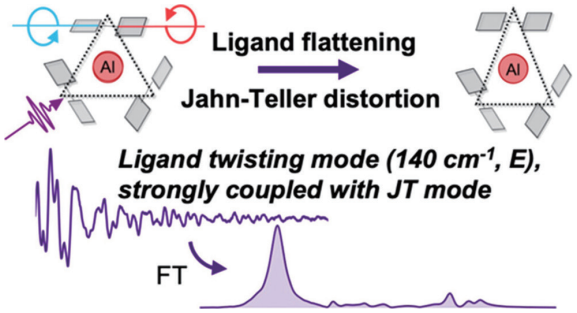

Figure 3. Schematic of the photoexcitation-triggered Jahn-Teller distortion in Al(III) dinuclear triple-helical complex.

3. Development of Two-Dimensional Fluorescence Excitation Correlation Spectroscopy

Polyatomic molecules in condensed phases undergo constant fluctuations in molecular structure and solvent environment. Fluorescence correlation spectroscopy (FCS) is advantageous in elucidating the fast fluctuation dynamics of freely diffusing molecules in solution, where a variety of chemical and biological processes occur. However, observing the fluctuation of diverse physical properties, such as electronic/vibrational spectra and ultrafast dynamics, still remains challenging. In this study, we developed fluorescence excitation cross-correlation spectroscopy for room-temperature solutions, which enables the study of spontaneous fluctuations in the excitation spectrum with microsecond time resolution. By employing Fourier transform spectroscopy with broadband femtosecond pulses and time-correlated single-photon counting, the method enables us to obtain an excitation wavelength-resolved fluorescence cross-correlation map in the microsecond to millisecond range, demonstrating the potential of this method to elucidate the transition between sub-ensembles in statistically equilibrium systems.

References

1) Y. Yoneda, T. Konishi, K. Suga, S. Saito and H. Kuramochi, *J. Am. Chem. Soc.* **147**, 12051 (2025).
2) T. Ehara, Y. Yoneda, T. Yoshida, T. Ogawa, Y. Konishi, T. Ono, A. Muranaka, H. Kuramochi, K. Miyata and K. Onda, *J. Am. Chem. Soc.* **147**, 26446 (2025).

Development of Designer Enzymes for Biomolecular Systems Engineering

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Life processes are sustained by a complex network of interconnected biochemical reactions. There has been a growing interest in re-engineering these biochemical reaction networks, which has implications for synthesis of chemicals and medical applications. We believe that the integration of unnatural chemical reactions, not found in nature but developed by humans, into this biochemical reaction network will pave the way for new ventures, leading to the production of various high-value-added compounds and the development of novel drugs with unique modes of action. With this ultimate objective in mind, our group is focusing on designer enzymes that catalyze unnatural chemical transformations. We are conducting a comprehensive study on the development of designer enzymes, drawing on coordination chemistry, catalytic chemistry, and protein engineering, as well as the development of technologies for their delivery into cells and organisms.

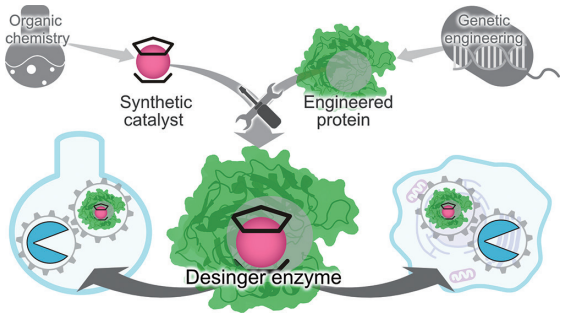

Figure 1. By combining synthetic catalysts developed through organic synthetic chemistry and proteins engineered through genetic optimization, we construct designer enzymes that possess non-natural functions. Using these designer enzymes, we aim to design chemical reaction networks within flasks and cells.

Selected Publications

- A. Ueno, F. Takida, T. Kita, T. Ishii, T. Himiyama, T. Mabuchi and Y. Okamoto, "A Cytokine-Based Designer Enzyme with an Abiological Multinuclear Metal Center Exhibits Intrinsic and Extrinsic Catalysis," *Nat. Commun.* **16**, 6781 (2025).
- Y. Okamoto, T. Mabuchi, K. Nakane, A. Ueno and S. Sato, "Switching Type I/Type II Reactions by Turning a Photoredox Catalyst into a Photo-Driven Artificial Metalloenzyme," *ACS Catal.* **13**, 4134–4141 (2023).
- H. J. Davis, D. Häussinger, T. R. Ward and Y. Okamoto, "A Visible-Light Promoted Amine Oxidation Catalyzed by a Cp* Ir Complex," *ChemCatChem* **12**, 4512–4516 (2020).
- Y. Okamoto, R. Kojima, F. Schwizer, E. Bartolami, T. Heinisch, S. Matile, M. Fussenegger and T. R. Ward, "A Cell-Penetrating Artificial Metalloenzyme Regulates a Gene Switch in a Designer Mammalian Cell," *Nat. Commun.* **9**, 1943 (2018).

1. Rational Design of a Synthetic Trinuclear Metal Complex Structures in a Protein Scaffold

Enzymes facilitate diverse chemical transformations in nature, with metal ions significantly expanding reaction capabilities. Examples include soluble methane monooxygenase (diiron enzyme for methane hydroxylation), nitrogenase (iron-molybdenum cofactor for nitrogen fixation), and photosystem II's oxygen-evolving complex (manganese-calcium cluster for water oxidation). In such metalloenzymes, protein scaffolds serve dual functions: Amino acid side chains act as ligands controlling metal ion reactivity, while defined internal spaces create reaction compartments enhancing rates and selectivity.

Designer metalloenzymes, created by incorporating synthetic molecules into proteins or constructing metal centers by using amino acid residues in protein, has proved the importance of reaction compartments by demonstrating enhanced reactivity and selectivity. However, using proteins as ligands lags behind their compartment applications due to difficulties in designing coordination chemistry at atomic levels. Designer mononuclear metalloenzyme development relies mainly on metal-substitution approaches and designing metal-binding site from scratch is further challenging. While some studies report construction of multinuclear metal centers using proteins and peptides as ligands, catalytically active examples remain limited with restricted scaffold variety.

Here, we successfully developed a designer enzyme containing a synthetic multinuclear metal complex structure by using proteins as the only coordination ligands.

As a model for grafting a multinuclear metal center into a protein scaffold, we have selected a synthetic trinuclear zinc complex (Figure 2a, b). This specific trinuclear zinc complex is a unique structural motif not typically found in natural enzymes. The choice of zinc ion was made due to its prevalence as one of the most abundant metal ions in biological systems.

For our study, we selected human macrophage migration inhibitory factor (MIF) as the scaffold protein because of its trimeric structure, which contains an internal pore suitable for hosting a synthetic trinuclear zinc complex structure (Figure 2c).

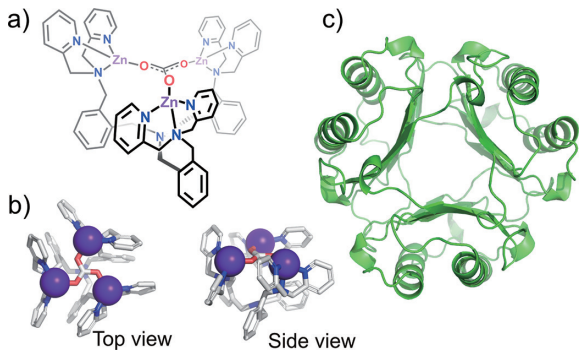

Figure 2. Building blocks used in this study. (a) Schematic and (b) crystal structures of the synthetic trinuclear zinc complex (CCDC 931956), and (c) crystal structure of human cytokine MIF (PDB code: 1MIF).

We have conducted a computational geometry search to identify suitable locations for placing histidine residues as ligands for the trinuclear zinc center. Subsequently, DFT calculations were performed to further refine the selection of candidate sites. As a result, we have prepared the identified variant, MIF(S61H/Y100H), along with an additional derivative, MIF(Y100H).

X-ray crystallography has verified the successful formation of the trinuclear zinc center in both MIF(S61H/Y100H) and MIF(Y100H) variants (Figure 3). The experimental structures align closely with the predictions from DFT calculations, illustrating the accuracy of our design strategy. Results from ITC analysis and DLS measurements confirm that the trimeric structures of the MIF variants are maintained in solution, with three zinc ions binding to the trimeric MIF variants.

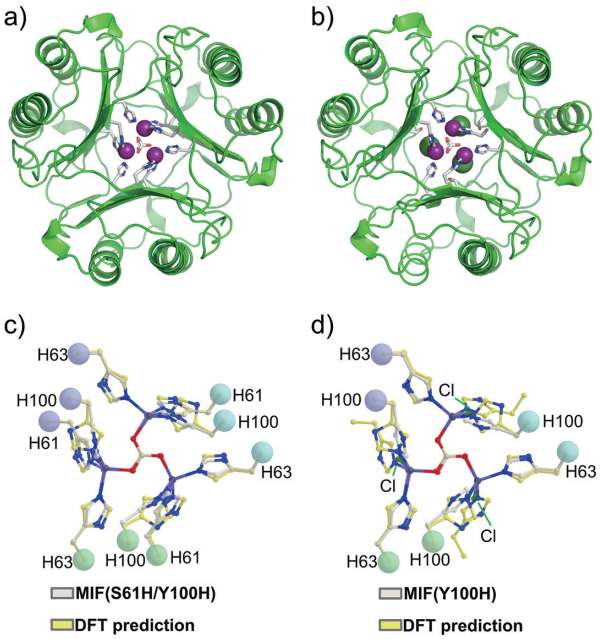

Figure 3. Crystal structures (a and b) of MIF(S61H/Y100H) (PDB code: 9JIZ; carbon: cyan) and MIF(Y100H) (PDB code: 9JJ0; carbon: cyan) in the presence of zinc ion. Superimposed images of the DFT-optimized trinuclear zinc center with (c) MIF(S61H/Y100H), and (d) MIF(Y100H) in the presence of zinc ion.

In the case of both MIF(S61H/Y100H) and MIF(Y100H) variants, hydrolytic activity was enhanced in the presence of zinc ions. This acceleration was not observed in the wild type MIF, indicating that the trinuclear zinc center plays a crucial role in catalysis. Interestingly, the trinuclear zinc center in the MIF(Y100H) variant exhibited higher activity compared to that in MIF(S61H/Y100H), achieving k_{cat}/K_M = 46.0 ± 0.4 M^{-1} s^{-1} at pH 7.9.

Reference

1) Ueno, F. Takida, T. Kita, T. Ishii, T. Himiyama, T. Mabuchi and Y. Okamoto, *Nat. Commun.* **16**, 6781 (2025).

Open up Future Electronics by Organic Molecules

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Keywords Organic Spintronics, Chirality, Organic Superconductor

Spintronics is a new ingredient of electronics in which a magnetic moment of an electron is utilized as an information carrier together with its charge. Spin-polarized current is one of the most important resources in spintronics, because it can drive devices such as ferromagnetic memory with spin angular momentum. In conventional spintronics, such a spin-polarized current is generated by passing a charge current through ferromagnetic metals. However, recently, researchers are finding other ways of spin-polarized current generation by using topological insulators and non-collinear antiferromagnets, which can sometimes be more efficient than those with ferromagnets.

Chiral molecules are attracting recent attention as a new source of spin-polarized current. Chirality-Induced Spin Selectivity (CISS) effect generates spin polarization parallel to or antiparallel to the electron's velocity depending on the handedness of the chiral molecule that is being passed through (Figure 1). Although the mechanism of CISS effect is still under debate, it seems to create spin-polarization higher than those of ferromagnets, which is surprisingly large when the small spin–orbit coupling energy of organic molecules is considered. In order to rationalize such a large effect, some microscopic hypotheses are proposed based on experimental results, whose proofs are being waited for. Our group is trying to unveil such mechanisms that drive CISS effect by using chiral crystalline materials.

The use of crystalline materials has several advantages. For example, one can employ theoretical framework with well-defined wave number of electrons. Another advantage is the size of the chiral material which allows direct attachment of detection electrodes in different positions. With these merits in mind, we are fabricating spintronic devices suitable for the CISS investigations.

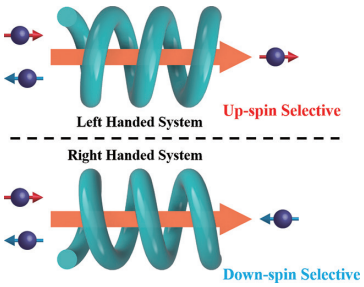

Figure 1. Conceptual schematic for CISS effect. P-helix molecule (lower panel) can transmit more electrons with spins antiparallel to the velocity (negative helicity electrons) than the other, while M-helix molecule (upper panel) favors transmission of electrons with parallel spin (positive helicity electrons).

Selected Publications

- R. Nakajima, D. Hirobe, G. Kawaguchi1, Y. Nabei1, T. Sato, T. Narushima, H. Okamoto and H. M. Yamamoto, *Nature* **613**, 479 (2023).
- Y. Nabei, D. Hirobe, Y. Shimamoto, K. Shiota, A. Inui, Y. Kousaka, Y. Togawa and H. M. Yamamoto, *Appl. Phys. Lett.* **117**, 052408 (2020).
- A. Inui, R. Aoki, Y. Nishiue, K. Shiota, Y. Kousaka, H. Shishido, D. Hirobe, M. Suda, J.-i. Ohe, J.-i. Kishine, H. M. Yamamoto and Y. Togawa, *Phys. Rev. Lett.* **124**, 166602 (2020).
- M. Suda, Y. Thathong, V. Promarak, H. Kojima, M. Nakamura, T. Shiraogawa, M. Ehara and H. M. Yamamoto, "Light-Driven Molecular Switch for Reconfigurable Spin Filters," *Nat. Commun.* **10**, 2455 (7 pages) (2019).

1. Spin Current Generation in a Chiral Organic Superconductor

Although *s*- and *d*-wave superconductors are in a spin singlet state at its ground state, a superconductor with broken mirror symmetry is expected to show spin triplet state when supercurrent is flowing, according to a theory developed by Edelstien.[1] This means spin polarization can be generated by applying supercurrent in a chiral superconductor. The magnetization direction that depends on the lattice symmetry has been recently calculated by group theory.[2] We have tested this idea by employing κ-(BEDT-TTF)$_2$Cu(NCS)$_2$ (hereafter, κ-NCS) which is an organic superconductor with chiral and polar crystal lattice. The space group of this crystal is $P2_1$, and its handedness is defined by the relative arrangement between the anionic Cu(NCS)$_2$ and cationic BEDT-TTF. This handedness can be experimentally determined by X-ray diffraction or circular dichroism (CD).

After confirming pure enantiomeric lattice system with CD microscope, a thin crystal of κ-NCS has been laminated onto a resin substrate with prepatterned gold and nickel electrodes. At temperature lower than superconducting T_c, an a.c. electrical excitation was applied to induce spin polarization (Figure 2). The spin polarization accumulated at the interface between κ-NCS and the magnetic electrode was detected as a built-up voltage that is dependent on the relative angle between the accumulated and ferromagnetic spins. We have compared the observed voltage with theoretical estimation and found that it exceeds the value predicted by Edelstein effect more than 1000 times. This surprising result suggests that there is a spin enhancement effect other than Edelstein effect, implying existence of an effect analogous to CISS for a chiral superconductor.

By measuring the angle dependency of this magneto–voltaic signal, the direction of accumulated spin could be determined. The observed spin polarization direction was dependent on the location of the detection electrode inside the crystal, and its arrangement was consistent with a magnetic monopole structure which has been hypothesized in a chiral molecule under non-equilibrium state with CISS effect. More specifically, the spin accumulation was forming an antiparallel pair on the upper and lower sides of the κ-NCS crystal. With a right-handed crystal, the accumulated spins showed outward spin pairs.

To our surprise, this spin accumulation could be observed in nonlocal measurements where the excitation and detection electrodes are separated by 600 μm. We have also fabricated a nonlocal detection device with a crystal possessing two chirality domains where right- and left-handed crystal structures are spatially separated. By exciting crystal domains at two different positions with opposite handednesses, we have observed a switching of antiparallel spin pairing mode from outward to inward. This corresponds to the sign reversal of magnetic monopole in the language of multipole expression.[3] An interesting point here is that the magnetic monopole is also breaking the mirror symmetry, and its sign is connected to the chirality of underlying crystal lattice, although the magnetic monopole is time-reversal-odd (*T*-odd chiral). Although this *T*-odd chirality is a metastable state and disappears at ground state, its relevance to the enantio-separation experiments in CISS effect is directly implied in this experiment. If one accepts the fact that a sign of such a metastable magnetic monopole at excitation can represent the sign of chirality (*T*-even electric toroidal monopole) in the lattice, both the large enhancement of spin polarization and the enantio-separation of chiral molecules at non-equilibrium state observed in CISS experiments can be naturally understood, because such a monopole can interact with magnetic substrate in a handedness-specific manner. Such an interaction will also provide a large exchange energy difference for each spin. In this sense, this experiment provides the first direct observation of antiparallel spin pair formation from coherent chiral system which seems to be connected to microscopic CISS mechanism. Although the Hamiltonians describing the chiral superconductor and chiral molecules are quite different, there are many common features such as singlet ground state, chiral lattice and quantum coherence over the entire body. Since the conversion from *T*-even spin current to *T*-odd spin accumulation requires time integration with an existence of spin reservoir, the spin carriers in chiral molecules and superconductors should be identified in future studies. We also expect emergence of superconducting spintronics once a sourcing of spin-polarized current in superconductor is established by chiral superconductors.
(BEDT-TTF = bis(ethylenedithio)tetrathiafulvalene)

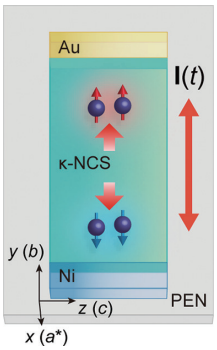

Figure 2. Device schematic for the detection of spin polarization in a chiral superconductor κ-NCS. By applying electrical current, electron spins are polarized along the current direction by CISS-like effect which can be detected as voltage across the κ-NCS/Ni interface. The amplitude of the signal is proportional to the accumulated spins at the interface.

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Design of Protein Functions Using Computational and Experimental Approaches

Research Center of Integrative Molecular Systems
Division of Trans-Hierarchical Molecular Systems

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Assistant Professor

Our research is to design a variety of protein functions using computational and experimental approaches. We try to (1) design enzymes from scratch and reveal the origin of the enzymatic activity, (2) control concerted functions by rationally engineering protein complexes and understand their mechanisms and (3) uncover roles of protein complexes in cells and control cellar functions by creating several customized proteins or protein complexes.

1. De Novo Design of ATPase

ATP hydrolysis plays pivotal roles in various proteins, including molecular motors and kinases. To elucidate the minimal structural requirements for ATP binding and hydrolysis, we computationally designed an ATPase from scratch, focusing the P-loop motif, a conserved phosphate-binding loop found in many naturally occurring ATPase.

Using computational design methods, we systematically explored an optimal topology that harbor the P-loop motif and facilitate binding to the adenine ring of ATP. Main-chain structures corresponding to the identified topology were generated, and amino acid sequences were designed to stabilize the main-chain structures and optimize ATP binding.

Biochemical assays for the designed proteins verified that one design was soluble, monomeric in solution, and exhibited ATP hydrolysis activity. Moreover, its crystal structure closely matched our design model and contained a P-loop motif with the typical features. We successfully demonstrated how to design a P-loop containing ATPase from scratch.

Reference

1) T. Kosugi, M. Tanabe and N. Koga, *Protein Sci.* **34**, e70132 (2025).

Award

KOSUGI, Takahiro; The Morino Foundation for Molecular Science (2025).