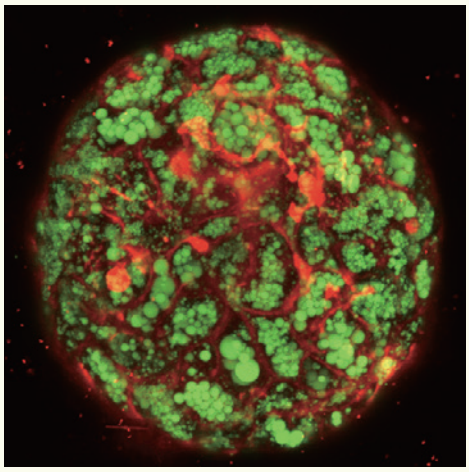
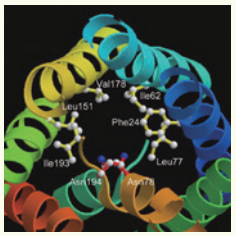
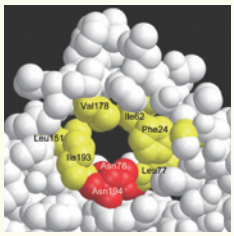
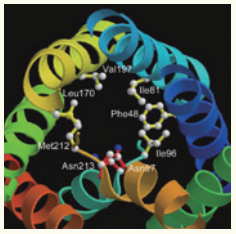
TMDU

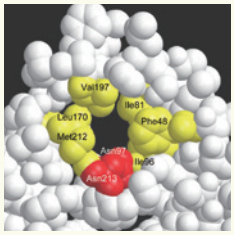
Research Activities 2020











TMDU-Committed to pioneering medical research

TMDU: Did you know...?

QS World University Rankings 2019 by Subject (Medicine and Dentistry)

	Medicine	Dentistry
National Rank	3	1
World Rank	51-100	10

SOURCE: QS World University Ranking by Subject 2019

THE World University Rankings by Subject 2020 (Clinical, Pre-clinical & Health)

Ranked #3 in Japan and #74 in the World

SOURCE: THE World University Ranking by Subject 2020

University Hospitals Promoting Our Research

	Beds	Outpatients Per Year
Medical Hospital	753	549,118
Dental Hospital	60	355,052

International Students

	No. of Int'l Students	No. of Countries
Graduate Schools	372*	34

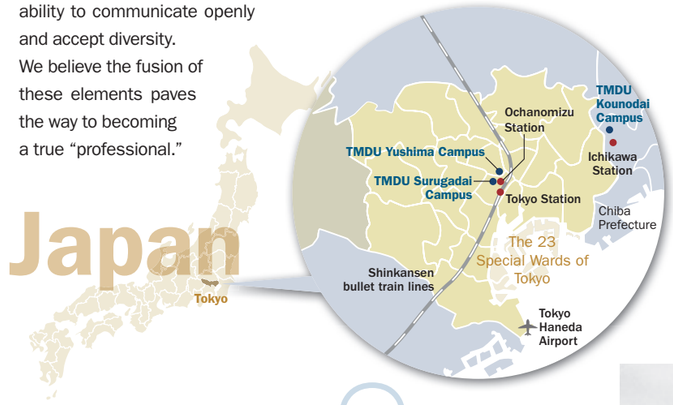
* About 21% of graduate school students are **International Students**

Contents

History and Location of TMDU

Standing at the sacred birthplace of scholarship in Japan

Tokyo Medical and Dental University was established as a national medical and dental educational institution on October 12, 1928. Currently, TMDU is located in the Yushima/Shoheizaka area of Tokyo, which is considered sacred ground for scholarship and learning in Japan. As Japan's only comprehensive medical university and graduate school, TMDU has provided advanced medical treatment through a fusion of the medical and dental fields. It has worked to cultivate professionals with knowledge and humanity, thereby contributing to human health and the well-being of society. The "knowledge" referred to here includes learning, technology, and self-identity, while "humanity" means culture, sensitivity, and the ability to communicate openly and accept diversity. We believe the fusion of these elements paves the way to becoming a true "professional."

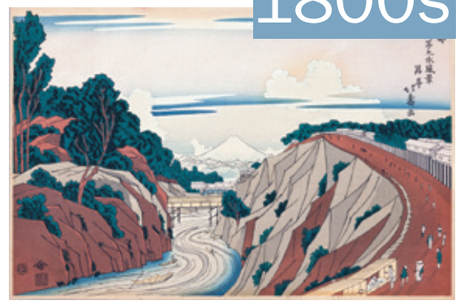


Today, TMDU is still located in Ochanomizu/Yushima district where its predecessor, the Tokyo National School of Dentistry, had moved in 1930, two years after its founding. TMDU has become known as one of the most excellent research universities in Japan.

TOKYO – The past and present

This landscape shows a view of Ochanomizu, where TMDU is located today. The buildings on the right-hand side, Yushima Seido and Shoheizaka School, were the center of scholarship since the 17th century, the Edo Period in Japan. Mt. Fuji can be seen in the far distance.

1800s



View of the Eastern Capital, Edo-Ochanomizu (woodblock by Shotei Hokuju)

1928

The Tokyo National School of Dentistry, the predecessor of TMDU, was established at Hitotsubashi.

2020

This monument at TMDU's Ochanomizu Gate commemorates the Birthplace of Modern Education. It honors Japan's modern education system, which was developed in this neighborhood after the Meiji Restoration, and marks TMDU's emergence at this site in 1930 as the world's first comprehensive medical-dental graduate school.

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TMDU establishes Medical Innovation Consortium

Toshihiro Tanaka

Director, Medical Innovation Consortium

Tokyo Medical and Dental University (TMDU) has developed the Life Course Consortium Concept, which identifies important research to be promoted and supports medical and dental studies that cover many aspects of human life. After establishing the 'Organ and Tissue Neogenesis' Consortium in 2017, the 'Medical Innovation' Consortium was launched in 2018.

Medical research that utilizes genomic information, including cancer genomic medicine, is one of the research fields in which the university excels, in both basic and clinical aspects. In April 2020, the M&D Data Science Center will be established as a research and education center for data science in the medical and dental fields. Data science is expected to further support genomic medical research.

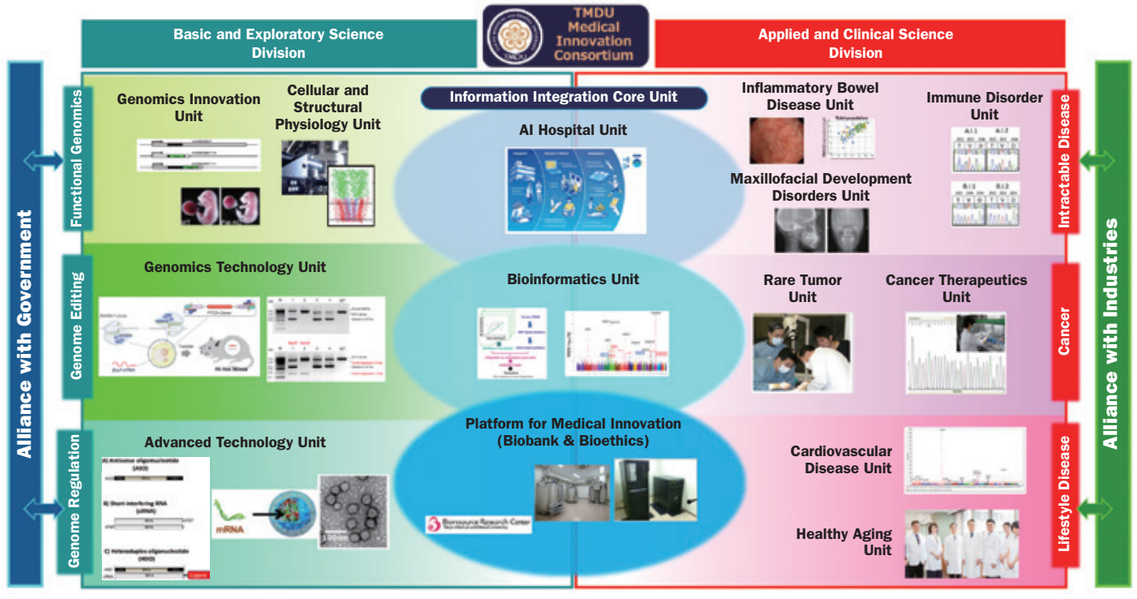
TMDU has strengths not only in genomic medical research and data science, but also in cell-structure physiology research using cryo-electron microscopy, drug discovery fields such as oligonucleotide and mRNA medicine, and efficient genome-editing technology. The 'Medical Innovation' Consortium has a mission to bring together these technological capabilities of the university and implement future medical technologies in society. A kick-off symposium was held on December 9, 2019.

The Medical Innovation Consortium consists of three organizations:

1 The Basic and Exploratory Science Division searches for undiscovered genome functions and develops new knowledge and technology.

2 The Applied and Clinical Science Division develops therapeutics for refractory diseases, cancer and lifestyle-related diseases.

3 The Information Integration Core Unit brings together multi-layered information for future medicine. It provides big-data analysis technology and processes high-quality biological samples and clinical information collected by the Bio Research Center. It also provides support on bioethical issues and acts as a bridge between the two other organizations in the Life Course Consortium.



Introducing the Units (Photos: Unit Leaders)

Basic and Exploratory Science Division

Genomics Innovation Unit

Fumitoshi Ishino

Professor, Department of Epigenetics

Contributing to future medicine through elucidation of human genome functions and discovery of disease-related genes

Cellular and Structural Physiology Unit

Yoshinori Fujiyoshi

Distinguished Professor, Cellular and Structural Physiology Laboratory (CeSPL), TMDU Advanced Research Institute (TMDU-ARIS)

Utilizing cryo-electron microscopy, structural and physiological studies available for future medicine

Genomics Technology Unit

Kohichi Tanaka

Professor, Department of Molecular Neuroscience

Promoting the development of genome-editing technology to support pathology elucidation and drug discovery

Advanced Technology Unit

Takanori Yokota

Professor, Department of Neurology and Neurological Science

Promoting the development of highly safe and practical drug discovery by heteroduplex oligonucleotide (HDO)/mRNA drugs

Information Integration Core Unit

AI Hospital Unit

Yoshikazu Nakajima

Professor, Department of Biomedical Information

Advanced integration and analysis of medical information using multidisciplinary artificial-intelligence collaboration

Bioinformatics Unit

Tatsuhiko Tsunoda

Professor, Department of Medical Science Mathematics

Contributing to future medicine by exploring disease-related genes and constructing prediction algorithms for precision medicine

Platform for Medical Innovation (Biobank & Bioethics)

Johji Inazawa

Professor, Department of Molecular Cytogenetics
Director, Bioresource Research Center

Contributing to future medical research through construction of the TMDU Biobank infrastructure

Applied and Clinical Science Division

Inflammatory Bowel Disease Unit

Mamoru Watanabe

Distinguished Professor, TMDU Advanced Research Institute (TMDU-ARIS)

Providing models of human healthy and diseased bowels by organoid, aiming for clinical application of innovative disease treatments

Immune Disorder Unit

Tomohiro Morio

Professor, Department of Pediatrics and Developmental Biology

Pioneering advanced research and personalized medicine in immune diseases by analyzing big data and establishing disease models

Maxillofacial Developmental Disorders Unit

Keiji Moriyama

Professor, Department of Maxillofacial Orthognathics

Analyzing genetic information on rare diseases that occur in the oral and maxillofacial regions to elucidate pathophysiology and develop new treatments

Rare Tumor Unit

Hiroyuki Harada

Professor, Department of Oral and Maxillofacial Surgery

Developing prophylaxis and new treatments for head and neck cancers based on a proven track record in medical and dental fields

Cancer Therapeutics Unit

Sadakatsu Ikeda

Associate Professor, Precision Cancer Medicine, Medical Hospital

Developing a foundation for creating real-world evidence, and contributing to the advancement of data science

Cardiovascular Disease Unit

Toshihiro Tanaka

Professor, Bioresouce Research Center

Developing precision medicine of cardiovascular diseases for a healthier super-aging society

Healthy Aging Unit

Kinya Ishikawa

Director, Professor, Center for Personalized Medicine for Healthy Aging

Analyzing genomic, lifestyle, environmental factors, etc., to provide total medical care that contributes to a longevity healthy society

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Structural physiology of membrane proteins by cryo-electron microscopy

Yoshinori Fujiyoshi

Distinguished Professor,
Cellular and Structural Physiology Laboratory (CeSPL),
TMDU Advanced Research Institute

Q: What motivated you to develop the cryo-electron microscope?

A: The starting point of my research came out of a simple quest to understand the molecular mechanisms that form cognitive ability and personality. Membrane proteins are important for the function of nerve cells, but lipid membranes are only about 5nm (50Å) in thickness. To analyze the molecular structures of such thin samples, electron beams are more suitable than X-rays, because their atomic scattering factors are larger. For this reason, I started to learn about electron microscopy.

The pioneers of electron crystallography are Richard Henderson and Nigel Unwin, and they first determined the structure of a membrane protein, bacteriorhodopsin (bR), at 7Å resolution in 1975. When I began researching electron microscopy in the late 1970s, I learned that biological samples could be severely damaged by the electron beams. Radiation damage of the biological samples made high-resolution analysis extremely difficult. After struggling through many attempts, we found that if the temperature was lowered to 8K or less, the damage to samples caused by electron beams could be reduced to about 1/20th compared with the damage at room temperature. Therefore, we set out to develop a cryo-electron microscope in 1983, and perfected a helium stage that could be built into an electron microscope in 1986. Since then, we have continued to make improvements and our eighth generation is currently under development. In 1997, using this type of cryo-electron microscope, we successfully analyzed the structure of bR at 3Å resolution.

Q: In electron crystallography, you have demonstrated the structure of many membrane proteins in addition to bR. What are the most impressive proteins in your research so far?

A: One example is the aquaporin (AQP) family of proteins, which are known to work as water channels. The AQP family proteins exist in all organisms, from bacteria to humans. A human has 13 types of AQPs that function in various parts of the body and are involved in numerous physiological processes.

An AQP1 channel passes 3 billion water molecules per second, but excludes any ions and even protons (H^+). If ions pass through a water channel, then ion channels cannot function, and if protons pass, the pH in the cell will change and cause dysfunction of the cell. However, since water molecules are connected by hydrogen bonds, protons could easily pass through the hydrogen bonds of a water wire formed in the channel. The mechanism of such rapid water permeation and high water-selectivity of AQP proteins gave us the puzzling questions.

In 2000, based on an analysis of the structure using electron crystallography, we proposed that there exists a hydrogen-bond isolation mechanism, which explained how AQP1 can block ion- and proton-permeation while maintaining rapid permeation of water molecules (Fig. 1). In 2009, we succeeded in individually observing eight water molecules in the channel of AQP4 (Fig. 2), and substantiating the proposed mechanism. This achievement was made possible by electron crystallography, which allows analysis of membrane proteins that are embedded in lipid membranes. The water molecules were not discriminated in the channel structure analyzed by X-ray crystallography even at higher resolution. This kind of counterintuitive notion could be attributed to the difference of surround atmosphere for structure analyses achieved in lipid bilayer or not. When X-ray crystallography is used to examine membrane proteins, the lipid membrane must usually be removed.

By the way, AQP2, which among AQPs plays an important role in the kidney, was discovered by Prof. Sei Sasaki, who was at that time a professor in the Department of Nephrology at TMDU (currently emeritus professor). Prof. Sasaki discovered the following mechanism: When a human is thirsty, a hormone called vasopressin is secreted from the postpituitary, and when it reaches the kidney, AQP2 in the cell comes to the cell surface and absorbs water, which otherwise would be excreted as urine. APQ2 inhibitors have potential for use in the treatment of heart failure and edema in cirrhosis, and we have started collaborative researches with Prof. Sasaki.

Q: Cryo-electron microscopy also plays an important role in single-particle analysis.

A: Yifan Cheng, who once was a postdoc in my lab, and his group analyzed the structure of the TRP (Transient Receptor Potential) channel at high resolution by single-particle analysis using a cryo-electron microscope and a high-performance camera that can detect electron beams directly with CMOS (complementary metal oxide semiconductor). That was in December 2013. Their work triggered an explosion in the study of structural biology by single-particle analysis.

In 2017, three scientists received the Nobel Prize for their work on the development of cryo-electron microscopy and single-particle analysis. One of them, Richard Henderson, explicitly cited our contributions at a press conference for the Nobel Prize when he said "Contributed people in Japan — Yoshi Fujiyoshi" (Fig. 3). The JEM-Z300CF cryo-electron microscope developed by JEOL Ltd. using my patent is starting to be widely used for single-particle analysis.

Q: You coined the term "structural physiology." What does this mean?

A: This term represents a discipline that seeks to understand physiological functions of membrane proteins from a structural point of view. For example, when we see an apple and judge that it is edible, then we reach out and bring it to our mouth — for each action, various molecules come into play, such as rhodopsin in the retina, ion channels in nerve cells, acetylcholine receptors at neuromuscular junctions, and so on. I have been attempting to determine the structures of all of these molecules and to understand human capabilities at the molecular structure level.

Three types of cryo-electron microscopy are available for studying three dimensional structures — electron crystallography, single-particle analysis and electron tomography. These are listed in order of the resolution they can achieve, but their order of importance in a biological sense might be the reverse. I've been conducting analysis using electron crystallography for many years; indeed, it took as long as 18 years in one case to analyze just one receptor. Therefore, recently we have also been using single-particle analysis, a technology in which there has been remarkable progress.

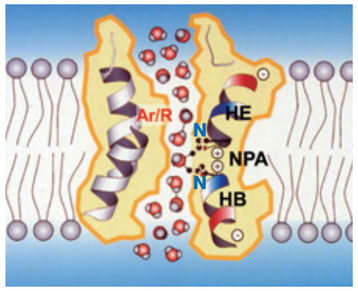

Fig. 1: Proposed hydrogen-bond isolation mechanism based on the structure of APQ1 at 3.8Å resolution. The middle of the channel is narrow enough to allow only one water molecule to pass. The two asparagine residues (N in the figure) form hydrogen bonds with the oxygen (red sphere) of the water molecule that has arrived, breaking the hydrogen bonds between the upper and lower water molecules.

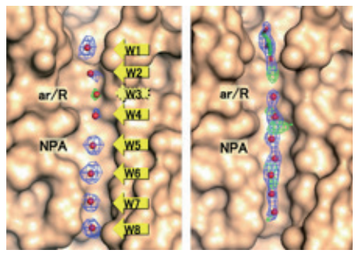

Fig. 2: APQ4 structure determined at 2.8Å resolution by electron crystallography (left), and structure analyzed at 1.8Å resolution by X-ray crystallography (right). Electron crystallography analyzed at lower resolution, but the eight water molecules (W1 to W8) in the channel are clearly discriminated unlike X-ray.

Fig. 3: Prof. Fujiyoshi (left) and Dr. Richard Henderson (MRC LMB) (right) at the Nobel Prize ceremony in Stockholm in 2017.

Currently, we are studying the mechanisms of acquired synaptogenesis, ion-channel gating and tight junctions formed by claudins, in addition to water channels. A typical example of single-particle analyses is structural analyses of gap-junction channels, which are the central feature of the electrical synapse. In a rather short period, we have been able to effectively analyze structures that are helping us understand the unique gating mechanism of the channel.

Q: Recently, you have been advocating the concept of "drug rescuing." Could you explain what this is?

A: Since around 2000, "evidence-based drug development." which identifies target proteins based on functional analyses of them and screens for their ligands, has become the mainstream method of drug discovery. However, even if promising compounds are found thorough screening, preclinical and clinical trials often encounter adverse effects and the success rate is as low as 1 in 30,000.

For this reason, "drug repositioning" to find other medicinal effects of already approved drugs is being actively pursued. However, there is a risk that the stores of these drug targets will be depleted. Also, in Japan, variations on the same compound will command the same price, so pharmaceutical companies do not make much profit from repositioning their drugs.

To overcome these problems, I came up with a strategy called "drug rescuing." The idea is to "rescue" compounds as well as drug target proteins that are discarded during the drug-development process. The promising way to implement this strategy is by determining a structure of the target protein binding the compound that was discarded. The structure will tell us detailed information about pharmacologic action, and allow us to improve the interaction of the compound. Importantly, the structure will also tell us the part of the compound that has no impact on binding. We can modify the compound at that specific location in various ways without affecting the optimized binding activity.

Drug rescuing is possible because single-particle analysis has accelerated structural analysis, but cryo-electron microscopes, which are essential for single-particle analysis, are expensive and difficult to maintain. For this reason, we have launched a business venture offering structural analysis services, and have started operation in cooperation with TMDU. Specifically, we will set up our equipment at Tokyo University of Agriculture and Technology as a hub center and plan to build remote operation systems in TMDU via fiber-optic connections. We will also conduct joint research with pharmaceutical companies and others, and hold workshops to share know-how for the method based on cryo-electron microscopy.

Q: Lastly, what are your future ambitions?

A: In addition to the contributions at TMDU I have already mentioned, I hope to use the network for other research groups as well as pharmaceutical companies. We have already succeeded in building up an effective remote operation system in TMDU, same of which could be installed in other groups.

There is another reason I founded the business venture: If we can be profitable enough, I would like to contribute to establishing an institute similar to The Medical Research Council, Laboratory of Molecular Biology (MRC LMB) in Cambridge, UK, where the number of Principal Investigators is small but already 19 researchers have been awarded the Nobel Prize. Although there are many reasons why MRCLMB is so successful, one important reason might be that the environment is conducive — a place where researchers can do their work without worrying about money. I therefore have a tremendous dream of creating a system that allows researchers to conduct research by using the income we will earn through venture-business sales and patent revenues.

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International collaboration reaps rewards in study of atrial fibrillation

Toshihiro Tanaka

Professor of Bioresource Research Center at TMDU

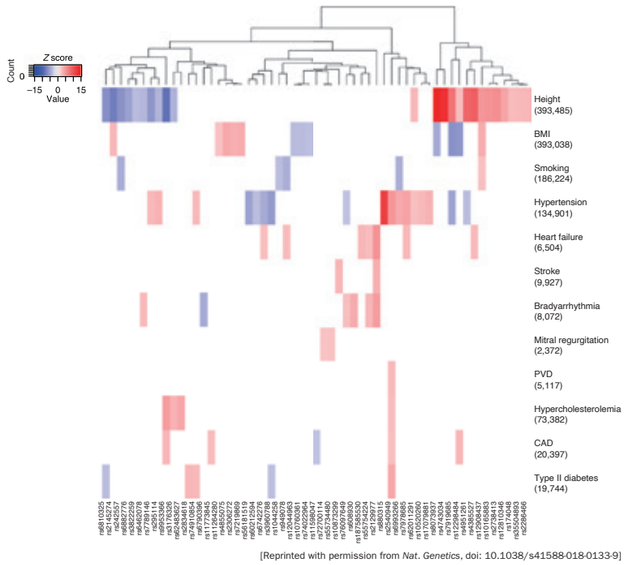
Q You have recently been involved in a large collaborative study of genetic loci associated with atrial fibrillation (AF). Please tell us about this work.

A: The study is the largest meta-analysis of genome-wide association studies of AF to date, and included more than half a million participants from 50 separate studies. The analysis identified 97 loci that were significantly associated with AF, 67 of which were novel. We were then able to link certain risk variants with candidate genes enriched within cardiac developmental, electrophysiological, contractile, and structural pathways, confirming their likely involvement with AF. Interestingly, many of the AF candidate genes encoded transcription factors, hinting at the complex, polygenic nature of AF.

Q You have previously carried out genome-wide association studies for AF, but the current study is a little different. Can you explain what sets it apart?

A: AF is a common heart rhythm disorder that significantly contributes to serious conditions such as heart failure and stroke. It is estimated that more than 33 million people are affected by AF worldwide. However, despite this global incidence, many association studies only focus on one, or perhaps several, ancestry groups. What sets the current study apart is the inclusion of patients from European, Japanese, African American, Brazilian, and Hispanic populations, and the fact that we performed both combined-ancestry and ancestry-specific meta-analyses. This approach is very important because while ancestry-specific loci were identified, we showed that the most common genetic susceptibility signals for AF were conserved across all populations, meaning that potential therapeutics need not be targeted to specific ancestry groups.

Cross-trait associations of AF risk variants with AF risk factors in the UK Biobank



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Q The work also includes a phenome-wide association study. Can you tell us a little more about this?

A: Phenome-wide association studies allow us to assess the pleiotropic effects of a disease-associated variant — essentially, whether a particular variant is also significantly associated with any other phenotypes. Using data from the UK BioBank, we showed that distinct clusters of variants were associated with both AF and height, body mass index, or hypertension. These findings may help us to better understand the diverse mechanisms involved in AF.

Q The study involved many international collaborators. How does this align with TMDU's research objectives?

A: Our study involved collaborators from 21 countries across Europe, North and South America, Australia, and Asia. TMDU has a strong focus on international collaboration and our work helps strengthen TMDU's relationships with leading research institutions around the world.

Dr. Tanaka graduated from the University of Tokyo, School of Medicine where he received his MD and PhD. He conducted research as Assistant Professor at the University of Tokyo from 1997 to 1999, then moved to RIKEN in 2000, and took the post of Deputy Director of RIKEN Center for Genomic Medicine in 2009. Since 2013, he has been Professor of the Research Division at the Bioresource Research Center of TMDU and Professor of Human Genetics and Disease Diversity at Graduate School of TMDU.

Nat. Genetics, **doi: 10.1038/s41588-018-0133-9**

Circulating microRNAs as an early indicator of atrial fibrillation

Tetsuo Sasano

Professor of Cardiovascular Medicine at TMDU

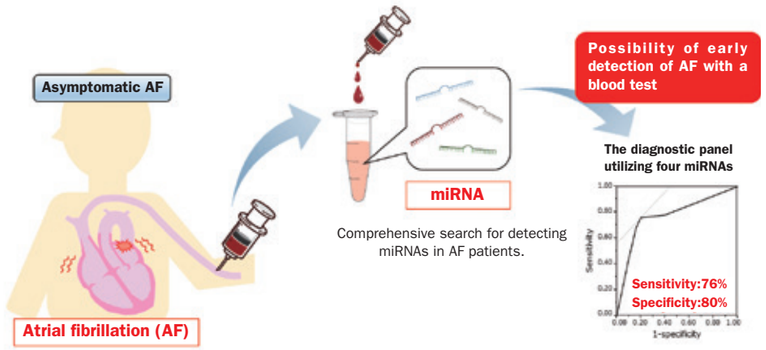
Q What made you focus on microRNAs (miRNAs) as biomarkers for atrial fibrillation (AF)?

A: These small, non-coding RNAs bind to target messenger RNAs and inhibit translation, thereby regulating gene expression. Recently, several studies have shown that miRNAs expressed in atrial tissue play a key role in the pathogenesis of AF. In addition, stable miRNAs have been detected in circulating blood. This led us to ask whether these miRNAs could be used as biomarkers for predicting AF.

Q Can you give us an overview of your latest findings?

A: We wanted to test our hypothesis that circulating miRNAs could be used as indicators of AF. To do this, we screened for 733 miRNAs in serum from patients with AF as well as 672 miRNAs in atrial tissues from mice with inducible atrial tachycardia. We compared the expression of the miRNAs in these patients and mice with that in suitable controls to identify miRNAs that were associated with disease. From this initial screening, we identified 11 miRNAs with significant changes in expression in both the patients with AF and the atrial tachycardia model mice. To confirm the association between the miRNAs and AF, we then quantified the expression of each of the candidate miRNAs in serum from 50 patients with AF and 50 healthy controls. This individual screening led us to identify four miRNAs that were upregulated in patients with AF. Encouragingly, the predictive accuracy of the four miRNAs as biomarkers for AF was very good, with a sensitivity of 76% and a specificity of 80%.

Prediction of AF with a blood test



Since AF strongly increases the risk of stroke, it is important to detect AF as early as possible, in order to start preventive therapy. However, about one-third of AF is asymptomatic, and it is sometimes difficult to detect AF in a case with paroxysmal AF. We screened circulating miRNAs in peripheral blood, and found four miRNAs associated with the presence of AF. The diagnostic panel utilizing these four miRNAs might be useful to predict an asymptomatic and/or undiagnosed AF with a blood test.

Q What are your next steps with this work?

A: Interestingly, only one of the miRNAs identified in our study has previously been directly linked to AF. While putative functions for the other three miRNAs have been suggested, the relationship between these miRNAs and AF is unclear. Therefore, we now hope to clarify the functional role of these miRNAs in relation to AF.

Q What are the clinical implications of your research?

A: AF is a risk factor for cardiogenic stroke, heart failure, and dementia but is often asymptomatic and can go undetected until the onset of serious illness. Therefore, the four miRNAs identified in our study could be developed into a diagnostic panel to predict asymptomatic AF via a simple blood test, allowing intervention before the onset of disease.

Dr. Sasano completed medical and graduate school at TMDU, where he received his MD and PhD. He became Associate Professor of Biofunctional Informatics at TMDU in 2011 and Professor of Cardiovascular Medicine there in 2019. His major research interests are arrhythmia, electrophysiology and gene therapy.

Circ J, **doi:10.1253/circj.CJ-17-1194**

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Microhomology: key to high-throughput mouse gene cassette knock-in

Kohichi Tanaka

Professor of Molecular Neuroscience at TMDU

Q How has available technology hampered the generation of genetically engineered knock-in mice?

A: Traditional CRISPR/Cas relies upon homologous recombination (HR) and the generation of vectors containing large regions of homology. This approach is cumbersome, time-consuming, and reaches efficiencies of only 10% – 20%, thus hindering large-scale application. We previously reported that the cloning-free CRISPR/Cas system facilitates HR-mediated gene cassette knock-in with efficiency of up to 50%. This is still lower than the approximate 100% efficiency observed for non-homologous end joining (NHEJ)-mediated gene knockout. Therefore, we sought to improve gene cassette knock-in efficiency in mammalian cells and zygotes.

Q Your new gene cassette knock-in method uses microhomology-mediated end joining (MMEJ). How does this improve the efficiency of gene cassette knock-in?

A: In mammalian cells, double-stranded DNA breaks (DSBs) are usually repaired by NHEJ, so the efficiency of any HR-based technique, such as CRISPR/Cas, will be low. MMEJ is an alternative pathway for DSB repair. Importantly for us, microhomologies are frequently found in the majority of CRISPR/Cas DSB-repair sites in mice. We harnessed this to develop the highly efficient and convenient CRISPR/Cas-based precision insertion into the target chromosome (PITCh) system.

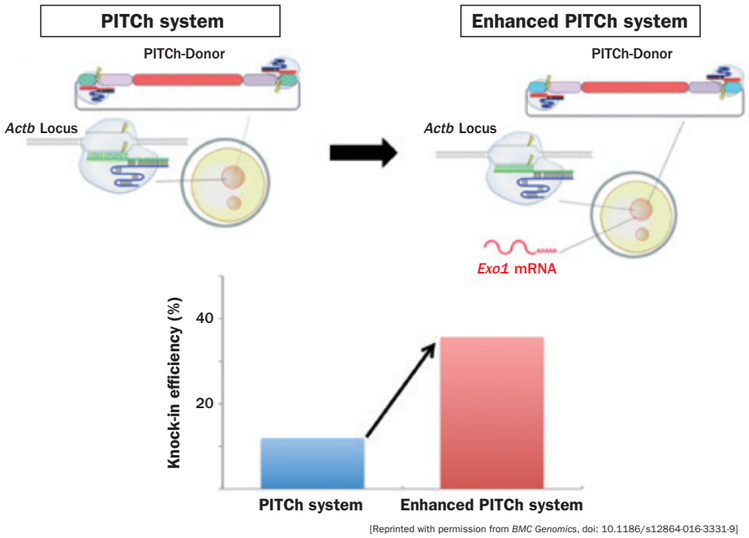
Q You were the first group to apply the PITCh system to mammalian cells. What does this mean for the generation of knock-in transgenic mice?

A: We performed a genetic screen to identify enhancers of MMEJ. Co-delivery of *Exo1*, involved in MMEJ, enhanced knock-in in human cells, and we applied this approach to generate knock-in mice. Using a combination of *Exo1*, PITCh, and the cloning-free CRISPR/Cas system increased the efficiency of knock-in mice to 30% from the approximate 10% achieved using PITCh and HR-based methods. The advantage of this approach over more traditional ones is the omission of laborious target vector construction.

Q What are the implications of this study for the field as a whole?

A: This approach requires a donor vector that can be generated using a single PCR, TA-cloning using primers conjugated with 40 bp of microhomology, and a genomic CRISPR RNA target sequence from any template plasmid containing marker or functional gene cassettes. This approach is exciting because it is scalable and can be used in a small research laboratory, or by a large consortium. This scalability, and the relative simplicity of the post-PCR TA-cloning, makes large-scale mouse knock-in projects feasible.

Generation of gene cassette knock-in mice by the enhanced PITCh system



[Reprinted with permission from *BMC Genomics*, doi: 10.1186/s12864-016-3331-9]

Dr. Tanaka received his MD and PhD from Niigata University. His postdoctoral research was performed with the Neural Network Team at RIKEN. He became a section chief of Neurodegenerative Diseases at the National Institute of Neuroscience in 1993 and assumed his present post at TMDU in 1998.

BMC Genomics, **doi: 10.1186/s12864-016-3331-9**

Novel oligonucleotide drug for gene therapy targeting neurological disease

Takanori Yokota

Professor of Neurology and Neurological Science at TMDU

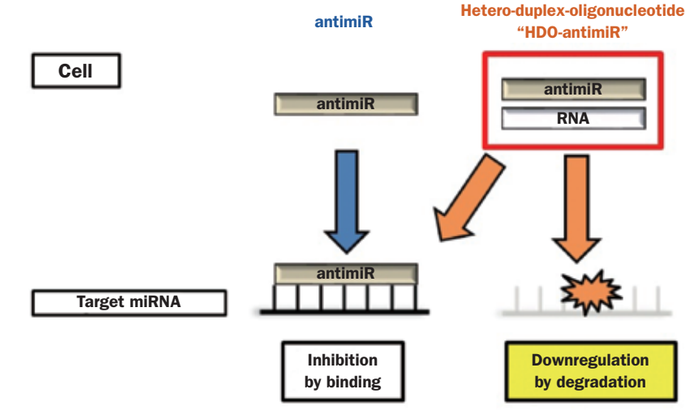
Q Dr. Yokota, you are Chairman of the Department of Neurology and Neurological Science at TMDU. Can you tell us about the key objectives and focus of the Department?

A: Given Japan's rapidly aging society, medical conditions that are associated with aging, such as stroke and Alzheimer's disease, have become a research focus. Innovations in molecular biology have enabled new possibilities for treating age-related neurological diseases. One of the main projects in our department is focused on the use of oligonucleotide drugs for gene therapy. Recently, we created a new class of oligonucleotide — DNA/RNA heteroduplex oligonucleotide (HDO) — which has great potential as a component of gene therapy.

Q What are the advantages of using HDO to treat neurological diseases?

A: The molecular structure and function of HDO are different from conventional genetic therapeutic approaches, and HDO has been found to be highly potent for molecular regulation. We were able to use HDO to develop a new form of a molecule that can inhibit specific types of microRNA (miRNA), which is implicated in various genetic diseases. This new type of anti-miRNA, or antimiR, may represent a new treatment approach for neurological disorders.

Intracellular mechanism of miRNA silencing by HDO-antimiR



This image illustrates the molecular mechanism for how double-stranded HDO-antimiR silences the targeted miRNA within cells in comparison to the original single-stranded antimiR.

Dr. Yokota received his MD from TMDU. His postdoctoral research was performed in several institutes including Tokyo Metropolitan Neurological Hospital and Sanford-Burnham Medical Research Institute in the United States. He returned to TMDU as Junior Associate Professor of Neurology in 2000, became Associate Professor of Neurology and Neurological Science in 2004, and assumed his present post in 2009.

Q What led you to focus on miRNA?

A: RNA plays an essential role in the regulation and expression of genes. As a type of RNA, miRNA is implicated in various cellular functions such as development, differentiation, growth, and metabolism. A number of human diseases may be caused by abnormal expression and organization of miRNA. Compounds that inhibit the activity of miRNA, or antimiRs, are being investigated for their potential utility in treating disease. Thus, we chose to focus on developing a new form of antimiR.

Q Can you tell us more about the new molecule that you developed?

A: We created HDO-antimiR, which has heightened efficacy against targeted miRNA and lower levels of toxicity in kidney than existing options. We found that HDO-antimiR was 12 times as efficient as conventional antimiR in terms of silencing targeted miRNA. Furthermore, HDO-antimiR has improved potency within cells. These improvements are due to the unique structure of HDO-antimiR, which has enabled it to behave differently from other types of miRNA inhibitors.

Q What are the clinical implications of your study?

A: Discovering new ways to silence malfunctioning miRNA could lead to new ways to treat diseases. In the future, HDO-antimiR technology may yield miRNA inhibitors for various diseases, including cancers, intractable neurological diseases such as Alzheimer's disease and Parkinson's disease, and cardiovascular disease, such as heart failure.

Nucleic Acids Res., **doi: 10.1093/nar/gkz492**

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Using nanoparticles to deliver cancer-killing microRNA

Johji Inazawa

Professor of Molecular Cytogenetics at TMDU

Q Dr. Inazawa, your work focuses on microRNAs as possible anticancer therapies. What are microRNAs, and how can they be used in cancer medicine?

A: MicroRNAs are short RNA molecules that are produced naturally in cells. Unlike messenger RNAs, which are used to synthesize proteins and are typically thousands of nucleotides long, microRNAs are non-coding and on average only around 20 nucleotides in length. MicroRNAs bind to and silence messenger RNA, which allows them to fine-tune different cellular processes by regulating gene expression. It turns out that many of these microRNAs regulate genes involved in cancer, and drugs that mimic microRNA activity may allow us to silence cancer-causing pathways in tumor cells.

Q You recently published a study looking at *miR-634* and its potential role in treating pancreatic cancer. Why did you focus on this particular microRNA and type of cancer?

A: We previously discovered that *miR-634* can act as a potent inducer of cell death in several different kinds of cancer cells. In our prior research, we had examined *miR-634* in only a handful of cancer cell lines. In the current study, we explored on a much larger scale, in over 100 different types of cancer cells. We saw that the microRNA had a powerful effect on multiple cell lines derived from pancreatic cancer. This naturally led us to think that *miR-634* could be key to developing a novel therapeutic agent for this cancer type.

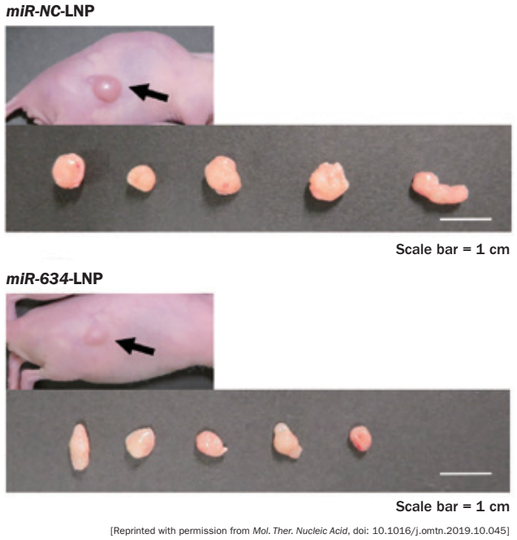
Q Can you describe the microRNA treatment and whether it was effective?

A: We used a lipid nanoparticle that allows us to deliver a mimic of *miR-634* through the bloodstream and into cancer cells. The mimic is structurally similar to *miR-634* and functions like the natural microRNA. We grafted pancreatic cancer cells into mice, then gave them microRNA treatment intravenously several times over a week. By day 21 we found that their tumors were, on average, less than half the size of those in mice given a control treatment.

Q What role do you think *miR-634* will play in the future of cancer therapy?

A: Therapies based on *miR-634* look to be quite feasible, not only for pancreatic cancer but for a variety of cancer types. Interestingly, *miR-634* exerts its effect on cell survival through the same pathways that confer chemoresistance, so it may even be possible to develop treatments for chemotherapy-resistant cancer. Our findings admittedly represent the earliest stages of drug development, but the initial results are very encouraging.

Representative images of pancreatic tumors removed from grafted mice on day 21 after treatment with a control microRNA (*miR-NC*-LNP) or therapeutic microRNA (*miR-634*-LNP)



[Reprinted with permission from *Mol. Ther. Nucleic Acid*, doi: 10.1016/j.omtn.2019.10.045]

Dr. Inazawa graduated from Kyoto Prefectural University of Medicine where he received his MD and PhD. He pursued postdoctoral research at Kyoto Prefectural University from 1982 to 1996, when he became Associate Professor at the University of Tokyo. He joined TMDU as Professor of Molecular Cytogenetics at the Medical Research Institute in 1998, and assumed the position of Director of the Bioresource Research Center in 2012.

Mol. Ther. Nucleic Acid,
doi: 10.1016/j.omtn.2019.10.045

Correlating circulating tumor DNA with patient prognosis in cancer

Sadakatsu Ikeda

Associate Professor of Precision Cancer Medicine, TMDU Medical Hospital

Q As Deputy Director of the TMDU Cancer Center, much of your work focuses on precision medicine. Can you explain what that means?

A: "Precision medicine" refers to personalizing medical treatment by using factors like genetics and molecular biomarkers in blood or tissue. It is sophisticated and, we hope, a more powerful way to treat disease. For example, cancer is traditionally diagnosed and treated based on where it originates in the body. But we're finding that the same molecular abnormality frequently underlies several cancer types, and that targeting abnormalities specific to each patient's cancer may be a more effective treatment approach.

Q One type of biomarker that has received attention lately is circulating tumor DNA (ctDNA). How can ctDNA personalize cancer care?

A: Circulating tumor DNA has enormous potential as a tool in cancer. As tumor cells die, they shed DNA, which enters the circulatory system and can theoretically be collected and analyzed using a simple, non-invasive blood draw. We are currently researching how ctDNA from these "liquid biopsies" can identify patients who are likely to have poorer outcomes.

Q Your team recently published an article describing this research. Can you elaborate on the study?

A: We looked at a gene called *MET,* which is commonly mutated or amplified in tumor cells. *MET* is an excellent biomarker candidate because it is associated with many types of cancer, and because there are treatments that target *MET*. We collected ctDNA from about 400 patients with different types of cancer, sequenced the DNA, and mapped it to a Real World Database, to identify *MET* abnormalities. We then reviewed the patients' medical charts to see if we could correlate their genetic findings to clinical findings.

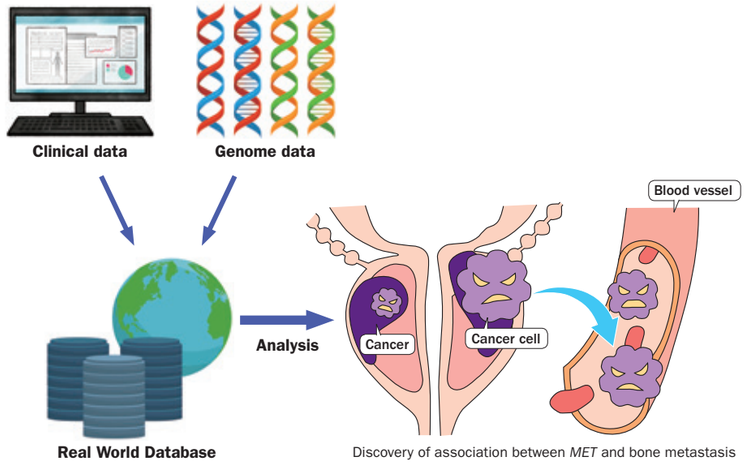
Q And what did you learn?

A: Our major finding was that patients with abnormal *MET* were more likely to have bone metastasis. Clinically, this nearly always means that the cancer has become incurable. In addition, these patients were more likely to have cancer-causing mutations in other genes, and a worse survival rate. We also found that the ctDNA analysis was sensitive — *MET* mutations were easier to detect with ctDNA than with DNA from a traditional tumor tissue biopsy.

Q What are the prospects for this method's use in precision medicine?

A: Our goal is to develop an assay to screen ctDNA with a blood draw, to help personalize the treatment plan for each patient. Reaching that goal is a long process, but our recent findings are an important step in that direction.

Utilizing patient data and Real World Database to predict patient prognosis



Discovery of association between *MET* and bone metastasis

Dr. Ikeda obtained his MD and PhD at Hokkaido University and performed his postdoctoral research at Harvard Medical School, Beth Israel Medical Center, the University of Michigan and The University of California, San Diego. He joined TMDU in 2016 and became Associate Professor of Precision Cancer Medicine at TMDU Medical Hospital.

Hematol. Oncol., **doi: 10.1186/s13045-018-0610-8**

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Uncovering the duality of *Porphyromonas gingivalis*

Toshihiko Suzuki

Professor of Bacterial Pathogenesis, Infection and Host Response at TMDU

Q You study the molecular mechanisms of bacterial infection and the host immune response. Why is it important to study the *Porphyromonas gingivalis* infection process?

A: *P. gingivalis* is one of the main bacterial causes of periodontitis. Its colonization of the oral cavity and ensuing chronic inflammation have been linked to the progression of systemic diseases such as cardiovascular disease and cancer, as well as inflammatory diseases such as rheumatoid arthritis. By studying the mechanisms of *P. gingivalis* infection, we may be able to identify potential targets for therapeutics that can halt the infection process.

Q Can you tell us about your findings?

A: *P. gingivalis* produces a range of proteases called gingipains that allow it to utilize small peptides in the periodontal tissue. Intriguingly though, conflicting reports also suggest that gingipains can both suppress the host inflammatory response and trigger inflammasome activation. Using wild-type and gingipain mutant strains, we attempted to unravel the seemingly contradictory effects of *P. gingivalis* on the host immune response. Our results confirmed that *P. gingivalis* infection triggers NLRP3-mediated inflammasome activation, inducing an immune response; however, activation was equivalent for all strains. Interestingly, heat-inactivated bacteria still induced a reaction, with secreted factors seemingly responsible for inflammasome activation. These findings are significant because they show that gingipains are not responsible for the *P. gingivalis*-induced inflammatory response.

Q Did you find any evidence that gingipains suppress the host immune response?

A: Yes, our results suggest that gingipains dampen the host's inflammatory response. *P. gingivalis* infection induces host-cell production of inflammatory cytokines such as IL-1β, IL-18, and TNF-α. Immature pro-IL-1β and pro-IL-18 are converted to their mature forms through proteolytic cleavage by activated caspase-1. However, immunoblotting and ELISA-based analyses of *P. gingivalis*-infected human monocytes showed that while mature IL-1β, activated caspase-1, and TNF-α were present in the supernatant of gingipain mutant-treated cell cultures, these fragments were absent from the wild-type *P. gingivalis*-treated cell cultures. Further analyses confirmed that gingipains proteolytically degrade the secreted cytokines and activated caspase-1, presumably suppressing the host immune response.

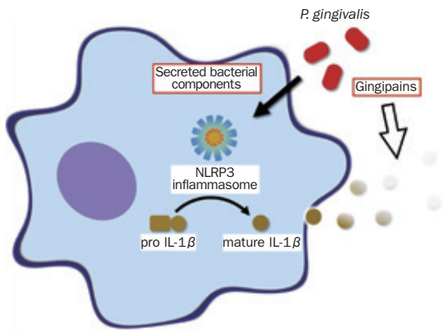
Q What are the future directions of your research?

A: The paradoxical effects of *P. gingivalis* in both triggering and suppressing the host immune response likely help it to stably colonize the periodontal tissues. However, we have yet to identify the secreted factors that trigger inflammasome activation. We now hope to characterize these factors at the molecular level to help us better understand the host-pathogen relationship.

Image of infected cells

Our image was selected as the cover art for the Dec. 4, 2018 issue of the *European Journal of Immunology*.

Paradoxical effects of *Porphyromonas gingivalis*



P. gingivalis triggers activation of NLRP3 inflammasome, resulting in the processing of IL-1β. On the other hand, it secretes gingipains that degrade IL-1β.

[Reprinted with permission from *Eur. J. Immunol.*, doi: 10.1002/eji.201847658]

Dr. Suzuki completed his graduate school at the University of Tokyo, where he received his Ph.D. He performed postdoctoral research at the Institute of Medical Science at the University of Tokyo. He became Professor at the University of the Ryukyus in 2006. He joined TMDU as Professor of Bacterial Pathogenesis, Infection and Host Response in 2015.

Eur. J. Immunol., **doi: 10.1002/eji.201847658**

Making multifunctional molecules

Takamitsu Hosoya

Professor of Chemical Bioscience at TMDU

Q Your research focus is on synthesizing multifunctional molecules. Please give us a brief overview of your latest publication.

A: Many biological analyses, including clinical tests, rely on multifunctional molecules to detect specific proteins, pathogens, or other target analytes. Usually, one end of the multifunctional molecule binds with the target, while the other attaches to a fluorescently tagged marker. There are advanced tests that require a "trifunctional" molecule with three binding sites in order to work correctly. However, adding a third functional group with conventional synthetic methods has proven to be very difficult, since the three groups need to be added sequentially, and usually require protection/deprotection reactions that are specific to that particular combination. Along with my TMDU colleagues, we were able to develop a new unified synthetic method for easily combining any three arbitrary functional groups in the same multifunctional molecule.

Q Describe this new method for creating trifunctional molecules.

A: We start by synthesizing a triazido platform molecule. Using three azido groups of different types — such as one sterically-hindered aromatic, one standard aromatic, and one aliphatic — we can easily perform orthogonal cycloaddition reactions to attach the three desired azidophilic ligands with high yields. Since this process is modular, we can even choose in which order to add the azidophiles. The final product contains the three chosen functional groups at the vertices of an essentially triangular-shaped multifunctional molecule. In the published research, we demonstrated our method by synthesizing a molecule containing a biotin linker, a HaloTag ligand, and a fluorescent BODIPY moiety.

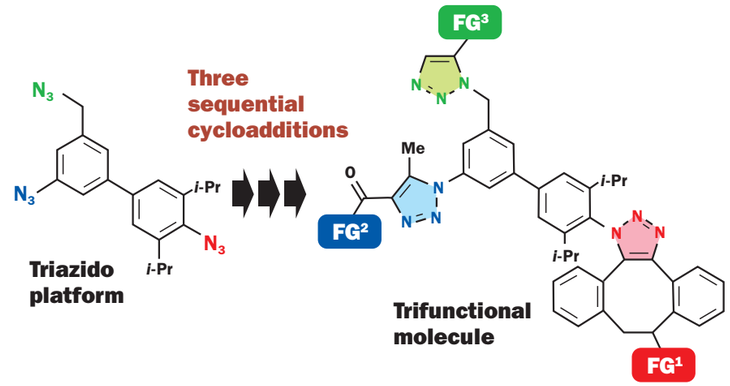
Q How does your research align with TMDU's focus areas?

A: We are very excited to contribute to TMDUs mission to provide new and better tools to clinicians and biologists to accelerate the delivery of rapid, accurate, and cost-effective health care diagnostic tests. We accomplish this by developing new synthetic methods to create a general platform for manufacturing multifunctional molecules. This advances TMDU's goal of enhancing public health and the quality of human life.

Q What are future directions for your research?

A: We hope to use this new method to prepare libraries of multifunctional molecules with various permutations of similar functional groups. Because we can now synthesize arbitrary combinations of groups with high yields, we can start applying them to the high-throughput screening of target analytes. This work may lead to new "point-of-care" rapid medical tests that can be performed cheaply with real-time results.

Combining three arbitrary functional groups



A facile strategy for the synthesis of trifunctional molecules involving three sequential selective triazole-forming reactions is proposed. This method exploits three kinds of mechanistically different azido-type-selective cycloadditions. Three different azidophiles could be efficiently connected to a triazido platform molecule with three types of azido groups in a consecutive manner, which rendered a practical trifunctional molecule readily available.

[Reprinted with permission from *Chem. Commun.*, doi: 10.1039/c8cc01195h]

Dr. Hosoya received his doctoral degree in Science at Keio University. From 1995 to 2005, he worked as Assistant Professor at Gifu University. He then worked as Associate Professor and Professor at Tokyo Institute of Technology. Since 2009, he has been Professor at TMDU. He also has been affiliated with RIKEN, and is now at the Center for Biosystems Dynamics Research there.

Chem. Commun., **doi: 10.1039/c8cc01195h**

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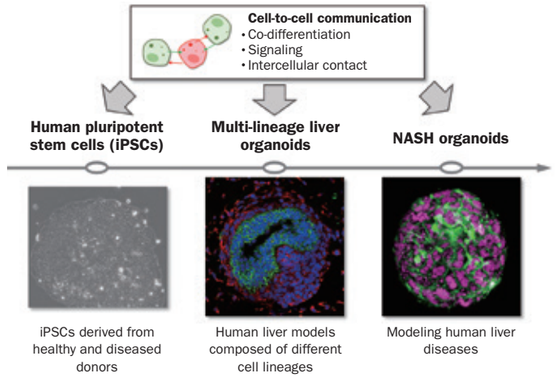
Toward understanding cell-to-cell communication using stem cell and organoid biology

Yosuke Yoneyama
Assistant Professor of Institute of Research at TMDU

I obtained my PhD in 2014 from the University of Tokyo where I completed my thesis work in Shin-Ichiro Takahashi's laboratory and then worked as a postdoctoral fellow there until 2018. During this training period, I focused on insulin-like growth factor (IGF), a peptide hormone that plays a critical role in body growth, metabolism, and aging of animals, including humans. Since IGF induces a variety of bioactivities, I was particularly interested in how IGF tightly controls its activities in target cells. Using molecular and cellular biology techniques, I found some key regulatory nodes in the signaling network that are spatially and temporally organized to control specific bioactivities.

In 2018, I joined Takanori Takebe's laboratory at TMDU as an assistant professor. Our laboratory is focusing on miniature organs *in vitro* called "organoids" that are derived from human stem cells, including induced pluripotent stem cells (iPSCs). Fortunately, I became part of a team studying non-alcoholic steatohepatitis (NASH), a liver disease that causes liver cirrhosis and hepatic carcinomas and is prevalent worldwide. We recently published research on a new human iPSC-derived organoid system that includes multiple lineages of liver cells and recapitulates the complex pathologies of NASH, such as steatosis, inflammation and fibrosis (*Cell Metab.*, doi: 10.1016/j.cmet.2019.05.007). By leveraging this organoid technique, we are trying to elucidate the molecular mechanisms underlying human NASH, focusing on cell-to-cell communication (hepatocytes, stromal cells, immune cells, endothelial cells, etc.), leading to the identification of potential drug targets for NASH. We are also tackling a novel form of cell-to-cell communication observed in pluripotent stem cells, which will eventually contribute to further understanding of human stem cell biology.

Stem cell & organoid technology for understanding cell-to-cell communication and diseases in humans



[Reprinted with permission from *Cell Metabolism*, doi: 10.1016/j.cmet.2019.05.007]

Elucidation of basophil functions in allergic inflammation

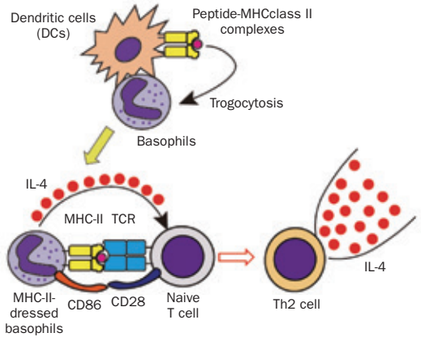
Kensuke Miyake
Specially Appointed Assistant Professor of Immune Regulation at TMDU

I started my research activities as I entered the advanced course for cultivating young researchers in 2012, when I was a fifth-year medical student at TMDU. After obtaining my MD in 2014, I directly entered the PhD course, and I obtained my PhD in 2017. I was then assigned to become the Specially Appointed Assistant Professor of Immune Regulation, and in 2019, was elected as a member of Next Generation Researchers at TMDU.

My research focuses on the functional roles of basophils, especially in the context of allergic inflammation. Basophils are the least common granulocyte, representing less than 1% of peripheral blood leukocytes. Research on basophil functions had been hampered by a lack of tools for investigating basophils. However, the recent development of several tools enables us to understand crucial roles of basophils in chronic allergic inflammation (*Allergol. Int.,* doi: 10.1016/j.alit.2017.04.007). Furthermore, basophils also modulate immune reactions by communicating with other immune cells, including T cells and macrophages. I first focused on the interaction between basophils and T cells, and revealed that basophils present antigens to naive T cells, after interacting with dendritic cells (DCs). Basophils acquire peptide-MHC class II complexes from DCs via a process called trogocytosis (*Proc. Natl. Acad. Sci. USA.*, doi: 10.1073/pnas.1615973114). Antigen presentation together with IL-4 produced by basophils lead to Th2 cell differentiation in the context of allergic inflammation. This work won many awards, including from the Japanese Society of Immunology, and was recommended by F1000 Prime.

In my current research, I focus on the interaction between basophils and macrophages. In the late phase of allergic reactions, basophils induce the differentiation of anti-inflammatory macrophages to suppress excess inflammation. I am now aiming to clarify the suppressive mechanisms of basophils by using transcriptome analysis of anti-inflammatory macrophages. I expect that the outcome of this research will produce novel targets for allergic diseases.

Basophils exert antigen presentation via trogocytosis-mediated acquisition of peptide-MHC class II complexes



Clinical research in geriatric dentistry for a super-aged society

Manabu Kanazawa
Junior Associate Professor of Gerodontology and Oral Rehabilitation at TMDU

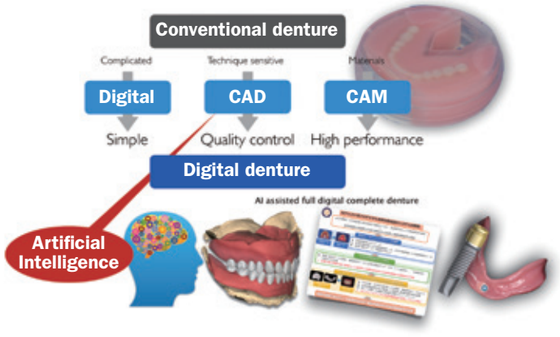
After graduating from TMDU in 2002, I received my PhD in 2006. I worked as clinical staff in TMDU Dental Hospital, and since 2008 as an assistant professor, I have mainly continued clinical studies on complete dentures (CD), implant overdentures (IOD) and computer-aided design/computer-aided manufacturing (CAD/CAM) dentures. In 2013-2014, I had the opportunity to move to McGill University in Canada to pursue research as a visiting professor of Oral Health and Society in the Faculty of Dentistry, under Prof. Jocelyne Fein, a world-famous professor in the field of IOD research.

My current research is focused on the clinical study of edentulous patients who use CDs, IODs and digital CDs. I have been interested in the outcomes and oral function reported by patients. Therefore, we have developed a chewing gum that changes color to assess masticatory performance (Masticatory Performance Evaluating Gum XYLITOL, Lotte) and acquired the patent in partnership with Lotte Corp. More recently, I have focused on nutrition while using a prosthesis, and reported on the importance of providing dietary advice to patients (*Clin. Nutr.*, doi: 10.1016/j.clnu.2017.07.022; *J. Prosthodont. Res.*, doi: 10.1016/j.jpor.2018.12.010). I also have some patents pending for digital CDs (*J. Prosthodont. Res.*, doi: 10.1016/j.jpor.2018.02.001), and am performing specific clinical research.

Additionally, I have signed an open-innovation agreement with Mitsui & Co., Ltd., to develop new artificial intelligence (AI) systems for use in dental diagnosis and treatment. This system also will be linked to studies in geriatric dentistry.

I hope to promote clinical studies based on higher quality evidence and to contribute to society by providing high quality specialized dental treatment.

The digital complete denture system for super-aged society



Analysis of age-related changes in skeletal muscle using progeria model mice

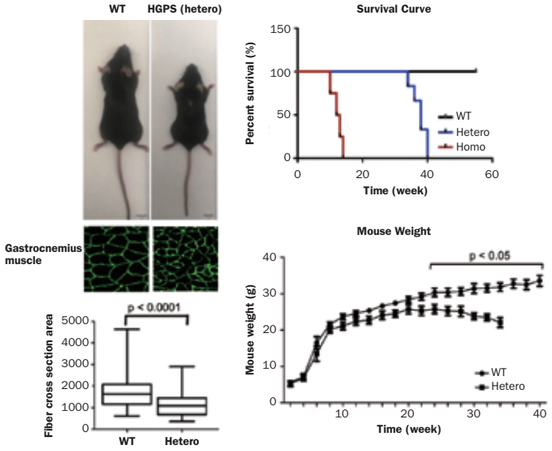
Kyoko Matsuzaki
Assistant Professor of Medical Biochemistry at TMDU

After receiving my PhD from the University of Tokyo, I worked as an assistant professor at the Institute of Medical Science at the University of Tokyo for six years. In 2015, I moved to the Department of Medical Biochemistry at TMDU.

Since I started my career as a researcher, I have been interested in cell biology and have analyzed cellular responses against various stimuli at a molecular level. I have focused particularly on cytoplasmic stress granules (SGs), a major adaptive defense mechanism, investigating the physiological function of SGs and revealing novel relationships between SGs and diseases. After moving to TMDU, I continued working on SG studies, and in 2018, we identified chemical compounds that suppress SG formation.

In my current position working with graduate students, I have broadened my interest to include analysis on an individual level. In particular, we are now focusing on senescence. To accelerate the research, we have established a knock-in model mouse of human Hutchinson-Gilford Progeria Syndrome (HGPS). HGPS is an autosomal dominant genetic disorder caused by mutations of *LMNA*, which encodes Lamin A, a nuclear membrane protein. HGPS patients have a heterozygous *LMNA* mutation and start exhibiting features of premature aging during childhood. Our hetero mice also showed a short life span and started to lose weight from around 24 weeks of age. Above all, the most notable feature in our hetero mice was their muscle atrophy. Thus, we are now focusing on this phenotype and working to figure out the molecular mechanism that induces age-related muscle atrophy.

Characteristics of HGPS model mice

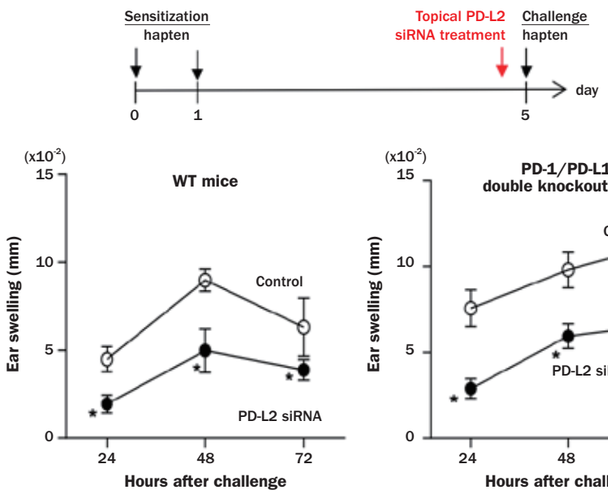


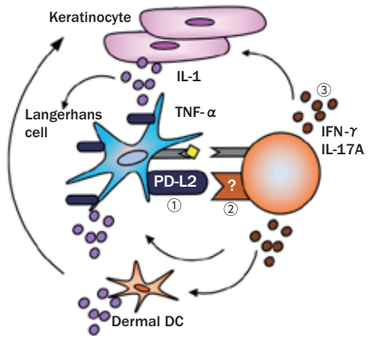
Nucleic acid ointment, the new contact-dermatitis cream

Allergic skin diseases, including contact dermatitis, affect millions worldwide. PD-L2 is a PD-1 immune-checkpoint ligand with unclear regulatory function. To understand the role of PD-L2, a team of TMDU researchers led by Emi Furusawa used small-interfering RNA (siRNA) to silence PD-L2 in a mouse model of contact hypersensitivity (CHS). PD-L2 expression was induced in epidermal Langerhans cells using a hapten antigen. Topical application of PD-L2 siRNA ointment inhibited the elicitation of CHS by suppressing early pro-inflammatory signals. Neutralization of PD-L2 protein using an anti-PD-L2 mAb produced similar results, showing that these results are PD-L2 specific. Topical PD-L2 siRNA also inhibited the development of CHS in mice lacking both PD-1 and PD-L1, indicating that this effect is PD-1 and PD-L1 independent. Although PD-L2 was thought to be an inhibitory molecule, these results show that PD-L2 may function as an activator in the elicitation phase of the CHS. It is expected that these results will lead to the development of PD-L2-targeted siRNA nucleic acid drugs for topical skin application.

J. Invest. Dermatol., **doi: 10.1016/j.jid.2019.02.037**

Topical PD-L2 siRNA treatment inhibits elicitation of contact hypersensitivity in PD-1-independent manner





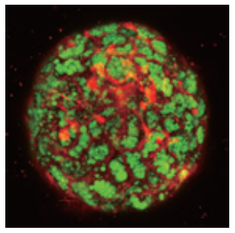
①Induction of PD-L2 in Langerhans cells (LCs)
②PD-L2 in LCs interacts with local memory T cells.
③Activated T cells secrete cytokines, resulting in the amplification of local inflammatory responses.

Inflamed 3D-miniature human livers open the way to drug discovery

Nonalcoholic steatohepatitis (NASH) is a common liver disease that may progress to cirrhosis or liver cancer. An effective treatment against the disease has not existed; highly therapeutic drugs have not been developed because of the lack of a culture system that recapitulates the pathology of steatohepatitis, especially inflammation and fibrosis. TMDU researcher Rie Ouchi and Professor Takanori Takebe's group, in collaboration with researchers from Cincinnati Children's Hospital Medical Center, have established the liver organoid model from human-induced pluripotent stem cells (iPSCs). The model includes immune cells that induce inflammation in liver, called the Kupffer cell, and stromal cells that are involved in fibrosis, called Stellate cells. The human liver organoid, including hepatocytes, Kupffer cells, and Stellate cells, accumulated fatty acids, including triglycerides with exposure of free fatty acid (FFA). The longer culture with FFA caused inflammation and fibrosis. The organoids from iPSCs derived from patients with congenital steatohepatitis, called Wolman disease, developed significant steatohepatitis phenotype after exposure to FFA. Importantly, the treatment of obeticholic acid and FGF19, which showed the effectiveness in clinical trials of NASH, alleviated the symptoms. This novel organoid technology may lead to the discovery of new effective drugs for steatohepatitis, including NASH, and moreover, become a platform for personalized treatment of the disease.

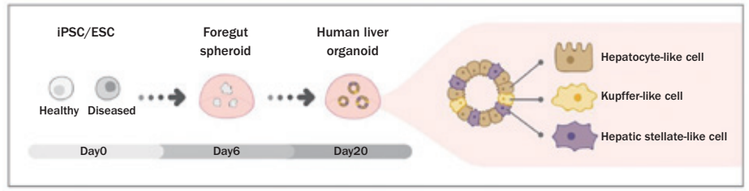
Cell Metabolism, **doi: 10.1016/j.cmet.2019.05.007**

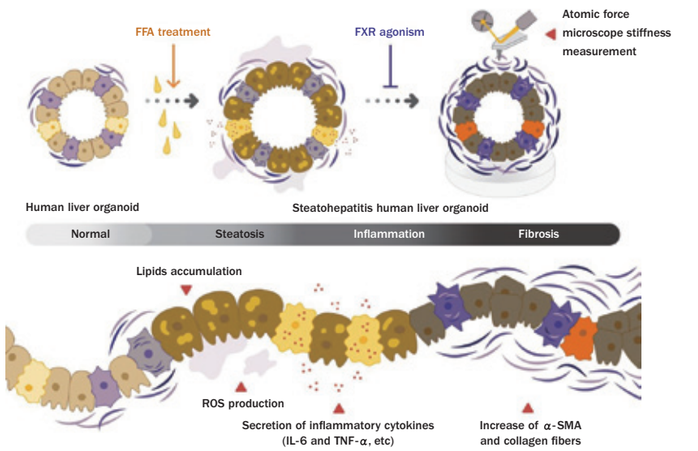
Confocal imaging of fatty liver organoid in a dish



Green indicates lipid accumulation; Red visualizes cellular membrane in a single organoid.

Generation of human fatty livers using custom-engineered induced pluripotent stem cells





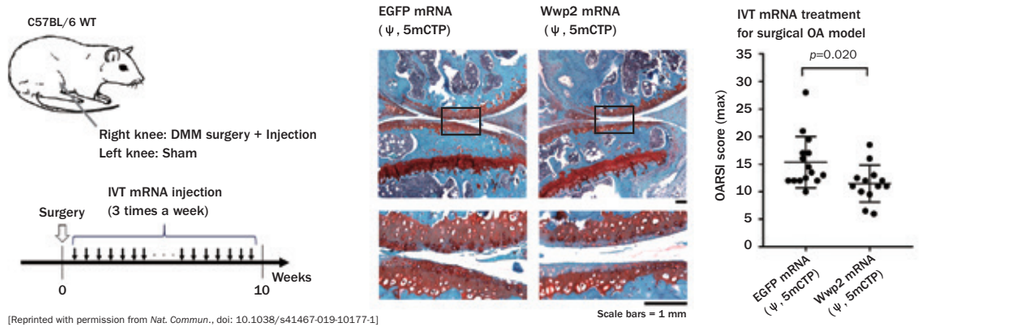
[Reprinted with permission from *Cell Metabolism*, doi: 10.1016/j.cmet.2019.05.007]

Wwp2 protects against cartilage damage in osteoarthritis

Osteoarthritis (OA) is a debilitating joint disease commonly seen in the elderly. The onset of OA involves the destruction of the articular cartilage, which is the tissue that forms the joint. Recently, it was discovered that the protein Wwp2 (WW domain-containing protein 2) is expressed abundantly in articular cartilage. Wwp2 is an ubiquitin E3 ligase, which functions to bind ubiquitin to a substrate. However, the role of Wwp2 in the articular cartilage remains unclear. To determine the function of Wwp2, an international team of researchers led by Hiroshi Asahara generated and examined Wwp2 gene-modified mice. Mice lacking Wwp2 and mice with an inactive Wwp2 showed increased symptoms of OA. Furthermore, when artificially synthesized mRNA (*in vitro* transcribed (IVT) mRNA) with the ability to produce Wwp2 in cells was introduced into the articular cartilage of mice, OA symptoms were reduced. The results from this study reveal that Wwp2 plays a role in protecting the articular cartilage from OA. Additionally, this may pave the way to new therapy options for treating OA.

Nat. Commun., **doi: 10.1038/s41467-019-10177-1**

IVT Wwp2 mRNA ameliorates articular cartilage destruction Injected mixture = IVT mRNA (ψ, 5mCTP) + invivofectamine + atelocollagen



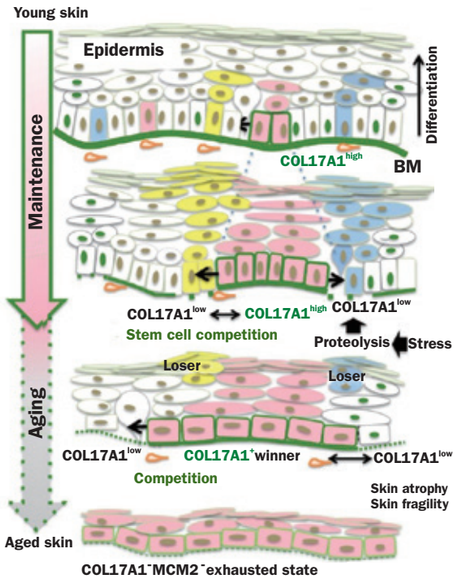
[Reprinted with permission from *Nat. Commun.*, doi: 10.1038/s41467-019-10177-1]

Skin vitality maintained through cellular "survival of the fittest"

Skin is thought to age when skin cells lose their ability to repair damage from environmental stress, but the connections between repair and aging are poorly understood. Researchers at TMDU, led by Emi Nishimura, investigated the dynamics of skin aging in mice and developed a model for how youthful skin persists over time. They found that epithelial stem cells respond to stress by breaking down collagen XVII (COL17A1), a protein that attaches cells to the membrane under the skin. As COL17A1-deficient cells easily detach from the membrane, adjacent COL17A1-rich cells outcompete them by multiplying and physically pushing them to the skin's surface for elimination. The selective competition processes keep skin "young" by continually replacing damaged cells. The researchers argue that skin ages when stem cells can no longer maintain high levels of COL17A and the pool of replacement cells is lost, and found that increasing cellular COL17A with topical chemicals ameliorates skin deterioration due to age. The discovery may lead to new restorative treatments for skin aging and age-associated diseases.

Nature,
doi: 10.1038/s41586-019-1085-7

Schematic of the epidermal aging program through COL17A1-mediated cell competition



COL17A1^{high} clones (winners) dominate in the basal interfollicular epidermis (IFE), whereas COL17A1^{low} clones (losers) are delaminated from the basal IFE during aging

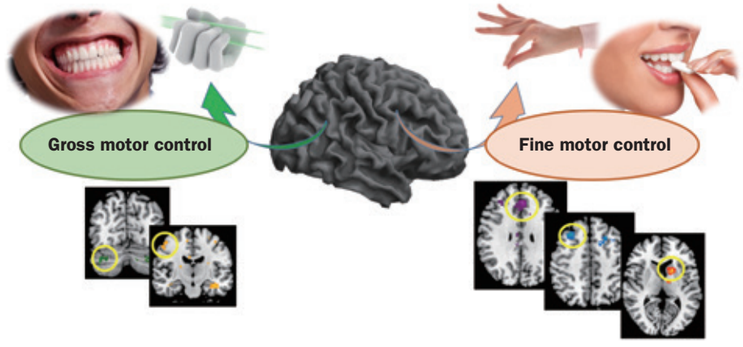
[Modified from *Nature*, doi: 10.1038/s41586-019-1085-7]

The brain and biting: brain activity correlates with incisal and molar bite force

Although biting is known to have a preventive effect against dementia, the relationship between biting and brain function remains largely unknown. The distinct functions of teeth (incisors for cutting and molars for grinding) are analogous to handgrip types (precision and power grips). Unlike handgrips, the brain correlates of fine and gross motor control during mastication have not been investigated. A research team led by Keiji Moriyama studied the motor-control systems engaged during incisal and molar biting. During molar biting, activity in motor function-related brain areas increased with increasing bite force, similar to the power grip. During incisal biting, activity in fine motor function-related areas decreased with increasing bite force levels, similar to precision grip. This indicates that the action of biting engages a single command system in the brain, but also engages two different movement control mechanisms. These findings elucidate the brain correlates of biting, and may help to elucidate the potential therapeutic effects of biting. Furthermore, our results may also help to clarify how occlusal hypofunction and dental treatments for it affect cortical motor-control systems.

Sci. Rep., **doi: 10.1038/s41598-019-44846-4**

Two different movement-control mechanisms in the brain are at work when biting an object

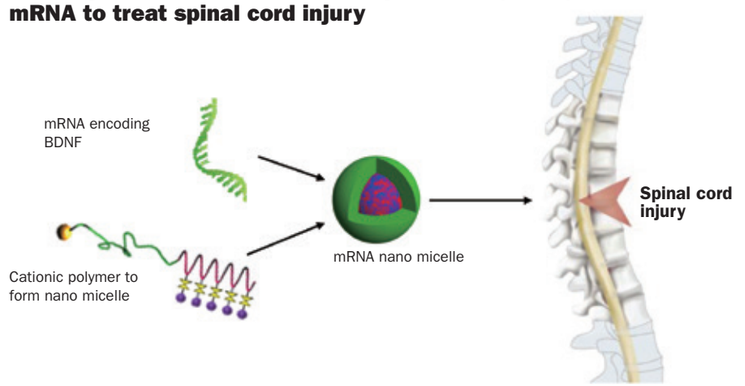


Delivery of BDNF mRNA enhances motor function following spinal cord injury in mice

"Secondary injury" in the days following spinal cord injury can impair motor recovery; therefore, finding ways to keep neural tissue alive during this critical period is key. Brain-derived neurotrophic factor (BDNF) is a secreted protein that promotes neuron survival. However, BDNF cannot cross the blood–brain barrier and its delivery requires frequent injections. While delivery of BDNF DNA can overcome these limitations, BDNF messenger RNA (mRNA) may be better yet; mRNA can transfect more cells and may produce more protein than DNA. To examine this, researchers at TMDU, led by Keiji Itaka, created a mouse model of spinal cord injury and administered BDNF mRNA to the injury site. mRNA-treated mice had doubled BDNF levels in spinal cord tissue (vs. no treatment) and an earlier motor recovery than non-treated and DNA-treated mice. This demonstrates the efficacy of BDNF mRNA treatment, which has implications for nerve function repair and regenerative therapy that does not require cell transplantation. mRNA drug treatments for central nervous system diseases and trauma are expected in the near future.

Mol. Ther. Nucleic Acids,
10.1016/j.omtn.2019.06.016

Delivery of brain-derived neurotropic factor (BDNF) mRNA to treat spinal cord injury

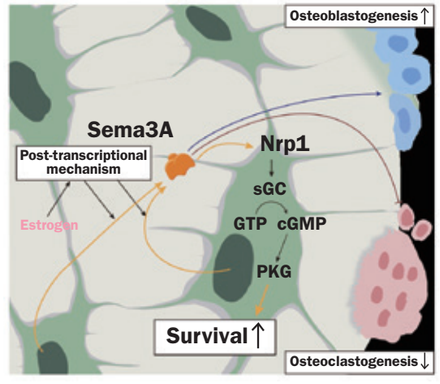


Activation of Sema3A signaling may protect against age-related osteoporosis

Osteoporosis is a disease characterized by reduced bone density and deterioration of the bone matrix, leading to fractures that can be physically disabling. Women are at a greater risk of developing osteoporosis as they age and reach menopause, when estrogen levels diminish significantly. While estrogen loss is known to cause the death of osteocytes needed to maintain bone homeostasis, the underlying mechanism is unclear. Tomoki Nakashima, leading a research team centered at TMDU, found that estrogen enhances the expression of an osteoprotective protein, semaphorin 3A (Sema3A), by suppressing microRNAs that target the 3' untranslated region of the Sema3A gene. They found that the Sema3A protein is secreted by osteocytes and promotes osteocyte survival through an autoregulatory loop. This loop was shown to operate through Sema3A acting on the sGC (soluble guanylate cyclase) signaling pathway. Importantly, the researchers were able to protect estrogen-deficient mice from bone loss by intravenously administering a small-molecule activator of sGC signaling. The findings suggest that targeting the Sema3A–sGC axis may be a viable therapeutic strategy to protect against bone loss due to aging.

Cell Metab.,
doi: 10.1016/j.cmet.2018.12.021

Autoregulatory loop of osteocytes via Sema3A



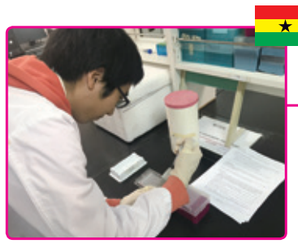
Estrogen induces expression of the osteoprotective protein semaphorin 3A (Sema3A), which acts on osteocytes to promote their survival and maintain bone homeostasis. An activator of soluble guanylate cyclase-cGMP signaling mimicked Sema3A action and ameliorated bone loss after ovariectomy.

Please contact us »» uraoffice.adm@tmd.ac.jp

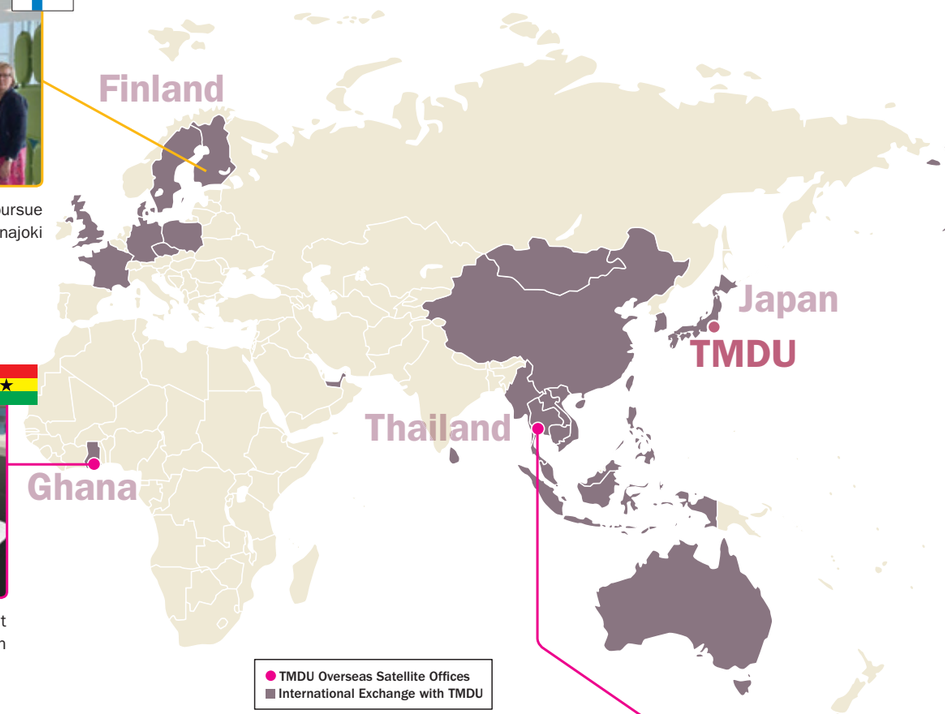
Sharing expertise and groundbreaking research around the world

Our international exchange activities in research and education are based in three centers, in Ghana, Thailand and Chile. We further promote educational collaboration with Harvard Medical School, Imperial College London and Australian National University. TMDU has 107 agreements in 31 countries in total.

Health Care Sciences majors pursue short-term training in Finland at Seinajoki University.



TMDU student conducts research at Ghana-TMDU Research Collaboration Center.



Dental students from TMDU study at Srinakharinwirot University in Thailand.

Japanese New Year's Festival attendees in the lobby of M&D Tower at TMDU.

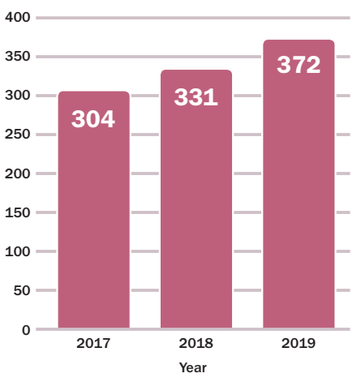
In fiscal year 2019 TMDU organized a total of ten events for international students. Two of them were hosted jointly with Juntendo University.

Students participating in the Summer Festival.

Students pursue clinical electives at Harvard Medical School in the U.S.

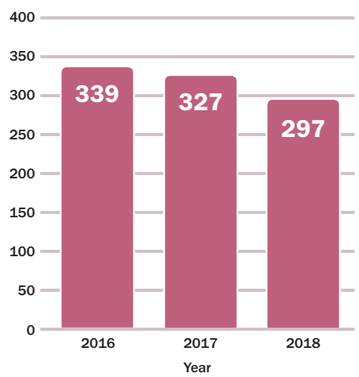
Project semester students from TMDU studied for six months at the University of Chile.

International Students



International students comprise about 21% of TMDU's postgraduate student body. May, 2019

Students Sent Overseas



About 20% of eligible students study abroad. March, 2018

How do you like life at TMDU?

Leila Nasiry Khanlar
(Iran)

My life as a PhD student has been busy but joyful, thanks to the friendly and collaborative atmosphere throughout the department. Aside from research, I have learned about personal interaction, teamwork, patience, and respect for others. Life outside the laboratory only becomes better with every passing day. I am grateful for this life-changing experience at TMDU, which has allowed me to meet and work with wonderful people.

Alapati Aimaitijiang
(Xinjiang Uyghur Autonomous Region, China)

Nothing is more attractive than studying at TMDU, where diversity is respected and you can communicate freely with people from all over the world. Although being away from one's home town and parents can be difficult, I found friendly support that helped me spend my first days on campus smoothly. TMDU provides us many challenging opportunities to immediately apply our newly learned skills and knowledge. I look forward to seeing how it also cultivates our independence and creative thinking.

Saleh Sherif Adel Abdelfattah
(Egypt)

I always thought that traveling gives you perspective and makes you grow as a person. That is especially true of my time at TMDU. Studying here continues to be a truly remarkable life experience. TMDU not only provides high-level education and equips students with up-to-date research skills, it also offers a global environment where you can learn about different cultures from all over the world. I believe this kind of experience prepares you for the highly connected world that we live in today.

TMDU is an outstanding environment for training and nurturing researchers for the future. I chose TMDU because of its high standards and qualifications, which are recognized around the world. My whole experience at this university has transformed me into a more confident and determined person. I will be eternally grateful to my dear mentors, who gave me tremendous hope and enthusiasm for pursuing cancer research.

Undrakh Ganbaatar
(Mongolia)

TMDU Research Activities 2020

Main campus of TMDU (Ochanomizu / Yushima District)

Cultivating professionals with knowledge and humanity, thereby contributing to people's well-being

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