FALL 2015

RIKEN RESEARCH

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PLANT STRESS
Keeping it under control

SHORT-LIVED NUCLEI
The atomic half-lives of exotic isotopes

THE CHICKEN IN THE EGG
Studies of early embryo development

GLOWING IN THE DARK

'Nano-lantern' proteins illuminate cellular processes without external light stimulation

Fourth RIKEN Cyclotron
RIKEN's fourth cyclotron launched in 1966 was 160 centimeters round and contributed to many advances in nuclear physics, chemistry, biology and medicine, until its decommissioning in 1990.

RIKEN, Japan's flagship research institute, conducts basic and applied experimental research in a wide range of science and technology fields including physics, chemistry, medical science, biology and engineering.

Initially established as a private research foundation in Tokyo in 1917, RIKEN became a national research and development institute in 2015.

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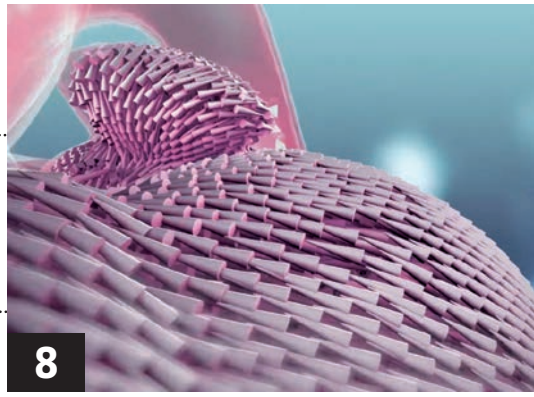

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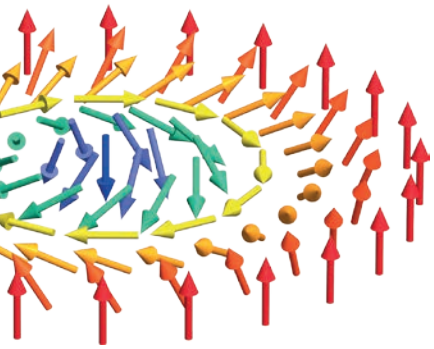

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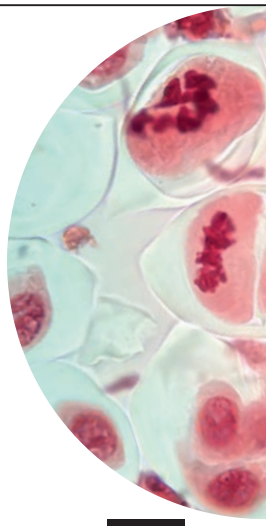
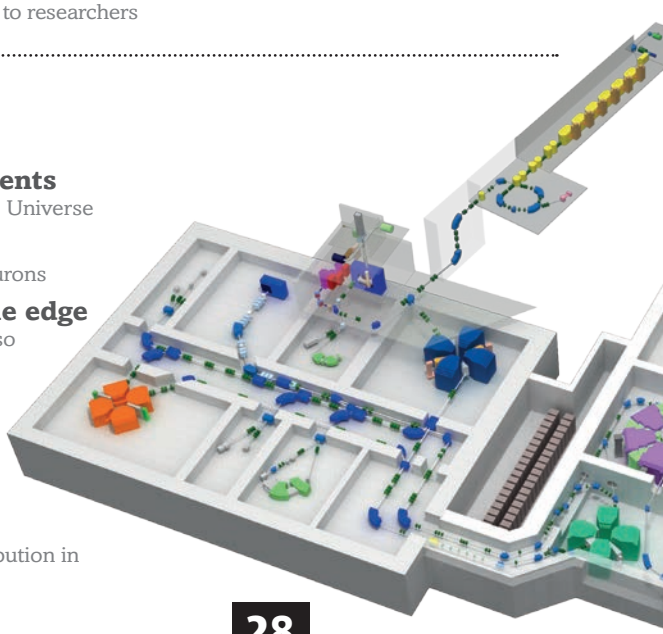
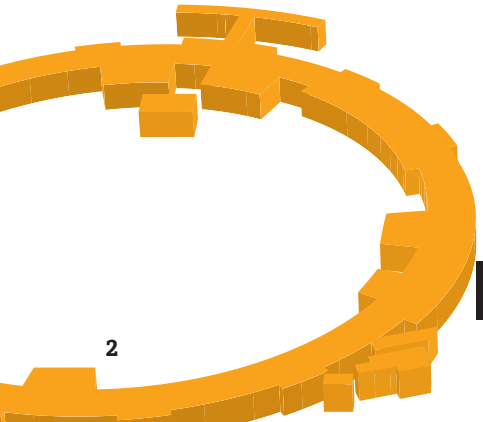






A season for excellence

Cover story: Researchers at the RIKEN Quantitative Biology Center have produced 'nano-lantern' proteins that can illuminate cellular processes without the need for external light stimulation. **Page 33**

Welcome to the Fall 2015 issue of *RIKEN RESEARCH*. Autumn is known in Japan as the season of harvest and fruit. It is a time when people can enjoy many delicious foods such as matsutake mushrooms, freshly harvested rice, chestnuts, salmon, sweet potatoes and persimmons, while admiring the beautiful autumn leaves.

It has been six months since Hiroshi Matsumoto became president of RIKEN and four new executive directors were appointed. On 22 May 2015, President Matsumoto announced a new management initiative called the RIKEN Initiative for Scientific Excellence. He has spent the last few months meeting with researchers across RIKEN to explain his strategy for fulfilling RIKEN's mission to deliver world-class results.

In this issue, the "Feature highlight" presents a study led by the director of the RIKEN Brain Science Institute, Susumu Tonegawa, and scientists at the RIKEN–MIT Center for Neural Circuit Genetics on the suppression of stress-induced depression simply by artificially reactivating memories stored during a positive experience. And our "Research highlights" report on lots of exciting new research, including three-dimensional and multilayered retinal tissue grown from embryonic stem cells and multicolored 'nano-lanterns' that can illuminate a range of biochemical processes without being excited by light.

Both "Perspectives" and "Places" articles feature the RIKEN SPring-8 Center, home to the SPring-8 Synchrotron Radiation Facility and the SPring-8 Angstrom Compact free electron LAser (SACLA). Director Tetsuya Ishikawa describes how researchers at the center are building x-ray sources that deliver brighter, shorter and more coherent x-ray pulses, which are accessible to all. And a fact-filled graphic reveals the number of papers that have been published based on research conducted at these facilities. We hope you will enjoy reading about the fruits of our labor.

Minding the brain in bipolar disorder

Tadafumi Kato

Deputy Director and Senior Team Leader

Molecular Dynamics of Mental Disorders, RIKEN Brain Science Institute

How did you become interested in neuroscience?

I was very interested in psychology and psychoanalysis in high school, and read books by Sigmund Freud on consciousness, preconsciousness and the role of dreams. I decided that I wanted to study humans from the perspective of a medical doctor, and so I joined the Faculty of Medicine at the University of Tokyo. There, I learned that the mind can be studied using neuroscience techniques, such as by measuring brain activity during rapid eye movement sleep when dreams typically occur. That was when I decided to focus on the brain as a means of understanding the mind.

How did you end up working on bipolar disorder?

After meeting several patients suffering from bipolar disorder, I was convinced that it was a disease of the brain. When bipolar patients are in their depressive state, they become immobile and cannot speak. After only one night of sleep deprivation, they suddenly go into a manic state of extreme excitement and hyperactivity. They look as if they are a different person. I felt that there must be a biological basis for such a dramatic behavioral change.

How has the diagnosis and treatment of bipolar disorder evolved in Japan?

When I first started my work 25 years ago, most psychiatrists focused on schizophrenia, because the symptoms of depression in bipolar disorder appeared to them as simple and not psychologically interesting. But the social burden of mood disorders like depression and bipolar disorder is much larger than schizophrenia, and the need for effective treatments even more important. I decided to write a few books to raise awareness among psychiatrists and the general public about bipolar disorder and reduce the stigma attached to it.

Unfortunately, not much has changed in terms of the methods used to diagnose bipolar disorder, and it is sometimes even misdiagnosed as schizophrenia.

I would like to show that human neuropsychiatric diseases can only be understood by combining basic and clinical science.

What has been your contribution to our understanding of bipolar disorder?

Researchers initially linked bipolar disorder to faulty intracellular signaling involving neurotransmitters. Our studies point to a role for mitochondrial dysfunction in bipolar disorder, which means that it is caused by more fundamental cellular dysfunction. We do not know, however, whether the neurons themselves are dying or whether their dendrites or axons are damaged.

What are your professional goals?

I would like to show that human neuropsychiatric diseases can only be understood by combining basic and clinical science. The natural sciences study objects and animals, whereas the social sciences study societies and people. Neuroscience lies between the two. Both basic science experiments using animal models such as mice and clinical science methods, which are more descriptive and observational, are needed to study the neural basis of mental disorders. In my work, I apply technologies from physics and chemistry to study the brain and understand the nature of human behavior and society. I hope to continue to bridge the research gap between basic and clinical science over the coming decades.

This is an edited excerpt of an interview between Tadafumi Kato and Ray Luo, a research scientist at the RIKEN Brain Science Institute.

Witnessing the birth of a star

Nami Sakai

Associate Chief Scientist

Star and Planet Formation Laboratory

Please describe your current research.
I study the formation of stars and surrounding protoplanetary disks of dense, rotating dust and gas using state-of-the-art radio telescopes from around the world. These early events in interstellar space eventually develop into the planets, asteroids and comets that make up a solar system. By analyzing the structural and chemical evolution of one of the most fundamental processes in the Universe, I hope to gain an understanding of the origins of our Solar System, and eventually, the origins of life on Earth.

> *I study the formation of stars and surrounding protoplanetary disks of dense, rotating dust and gas...to gain an understanding of the origins of our Solar System.*

How did you become interested in your current field of research?
When I was in elementary school, my father showed me the craters on the Moon through a telescope. That inspired me to study outer space and find out how stars are born. I decided to major in physics and joined a laboratory that focused on stellar nurseries, known as molecular clouds, and their chemical evolution. That was when I set myself a concrete goal of learning about the chemical origins of the Solar System, and whether it is unique or universal. I also hope to nurture young researchers who will continue to answer these questions for years to come.

What excites you the most about your current research?
There are so many things that we still do not understand—this is true of almost every field, but especially of astronomy. Our research typically involves making predictions about cosmic phenomena and then using telescopes to verify these predictions. More often than not, the results are completely unexpected. When that happens, it is exciting to realize that you are the only person in the world to know the unexpected nature of the Universe at that moment in time, and to imagine what effect it will have, if it turns out to be true, on subsequent research.

What has been the most interesting discovery in your field in recent years?
The discovery of hollow molecules of carbon called fullerenes in space, including spherical buckyballs (C_{60}), though this finding has had little bearing on my own research.

How has being at RIKEN helped your research?
I joined RIKEN in April 2015 as an associate chief scientist. Unlike at a university, we don't have any teaching obligations at RIKEN and can therefore concentrate on our research. We can conduct our research freely, without being restricted by the boundaries between different research fields, such as physics and chemistry. One day, for example, I happened to run into an old friend of mine from my high school days who is a researcher on the same campus at Wako city, near Tokyo. Even though he came from a different research field, we started conducting joint seminars, which made me realize how interdisciplinary RIKEN really is.

How do you balance family life with your work at RIKEN?
Whenever possible, I avoid working late and on holidays, and I try to spend time with my family. I don't have many rigidly scheduled commitments such as classes, so it is relatively easy for me to take time off if my son were to come down with a fever, for example. ■

Careers at RIKEN

For further information, visit our Careers page:
Website: www.riken.jp/en/careers
E-mail: pr@riken.jp

Briefs

World Scout Jamboree

RIKEN ran a scientific workshop at the 23rd World Scout Jamboree held in Kirarahama nature park in Japan's southern prefecture of Yamaguchi from 28 July to 8 August 2015. More than 33,000 Scouts aged 14 to 17 and their leaders from 152 countries and regions attended the event organized by the World Organization of the Scout Movement every four years.

This year's theme—WA: A spirit of unity—derives from the Japanese character *wa*, which epitomizes unity, harmony, cooperation, friendship and peace. Participants learned about global problems, different cultures and the natural environment. "We hope that the young Scouts will take their once-in-a-lifetime experience to their countries and mature into citizens of the world who contribute to solving global problems," said Yasushi Watanabe at the RIKEN Nishina Center for Accelerator-Based Science.

"The workshop was designed to nurture an interest in science and technology among young Scouts," said Atsushi Taketani from the Neutron Beam Technology Team at the RIKEN Center for Advanced Photonics (RAP). Scouts learned about the nature of light by making spectrometers and light-emitting diode lamps. "They understood very well that there are a variety of types of light and that light is important for the development of agriculture and industry, medicine and communications, as well as for art and culture," said Chiko Otani of the Terahertz-wave Research Group, RAP.

www.23wsj.jp/index_e.html

Initiating scientific excellence

RIKEN President Hiroshi Matsumoto announced a five-point strategy for scientific excellence at a press conference on 22 May 2015. The RIKEN Initiative for Scientific Excellence is designed to fulfill RIKEN's mission as a comprehensive research institution to generate world-class results. The strategy will help to ensure that Japanese innovation promotes coexistence with nature, contributes to the progress of humanity and sustains one of the world's leading economies. "We will work with Japan's universities to enhance Japan's scientific prowess, serve as a science and technology hub for research institutions and industries around the world, and achieve world-class results grounded in high ethical standards," states the text of the initiative.

The initiative's first goal is to pioneer a research management system that will serve as a model for all national research and development institutes in Japan. The system includes increasing the responsibilities of RIKEN headquarters, integrating personnel systems for permanent and fixed-term employees and introducing a tenure-track system. The strategy further outlines RIKEN's plans to lead the world in achieving new research and development results through scientific excellence, and become a hub for science and technology innovation as well as a focal point for global brain circulation. Finally, RIKEN promises to foster the development of world-class leaders in scientific research through the design and implementation of a long-term, stable employment system offering attractive career paths for young researchers of superior ability.

www.riken.jp/en/pr/topics/2015/20150522_3/

RIKEN President Hiroshi Matsumoto and Executive Director Sawako Hanyu at a press conference announcing the launch of a new initiative for scientific excellence.

Noyori named RIKEN Fellow

The previous president of RIKEN, Ryoji Noyori, was awarded the title of RIKEN Fellow at a ceremony on 16 June 2015 at RIKEN's Wako campus. The distinction is intended to honor individuals with an impressive record of scientific achievements who can contribute to the further development of RIKEN. It has been conferred on only one other person—RIKEN Brain Science Institute Director Susumu Tonegawa.

In presenting the award, President Matsumoto praised Noyori for his leadership of RIKEN from October 2003 to March 2015 and highlighted his many achievements. Most notably, Noyori's tenure saw the successful completion of two major infrastructure projects—the K computer and the SPring-8 Angstrom Compact free electron LAser (SACLA).

In his address, Noyori recalled some of his term's highlights—the visit by Emperor Akihito and Empress Michiko of Japan in 2006, the discovery of element 113 in 2012 and the recent launch of the first clinical trials using induced pluripotent stem cells. Noyori spoke about how he looks forward to seeing RIKEN grow under Matsumoto's RIKEN Initiative for Scientific Excellence and to celebrating the institution's centennial in 2017.

www.riken.jp/en/pr/topics/2015/20150626_1/

Germany–Japan chemical biology symposium

The fourth joint symposium of the RIKEN–Max Planck Joint Research Center for Systems Chemical Biology was held on 11–14 May 2015 in Kobe, Japan. Over 70 researchers presented their results at the annual event, which Germany and Japan take turns to host.

The joint research center was established in 2011 by RIKEN and the Max Planck Society to promote collaborative research and become a leading global institution in the field of systems chemical biology. Its core members include center director, Hiroyuki Osada, and group director, Naoyuki Taniguchi, from the RIKEN Global Research Cluster, as well as Herbert Waldmann, a director of the Max Planck Institute of Molecular Physiology, and Peter Seeberger, a director of the Max Planck Institute of Colloids and Interfaces.

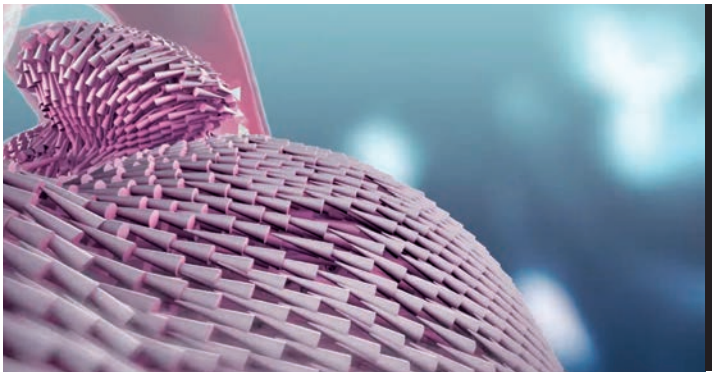
On 11 May, Kohei Tamao, director of the RIKEN Global Research Cluster, welcomed participants to the symposium and then there was a greeting from Ingo Karsten, consulate-general of the Federal Republic of Germany, Osaka-Kobe. The speakers touched on the three-decade history of collaboration between Japan and Germany and expressed high hopes for the center. Their introductory remarks were followed by a keynote address from Kazuo Shinozaki, director of the RIKEN Center for Sustainable Resource Science.

www.riken.jp/en/pr/topics/2015/20150518_1/

Prize-winning heartbeats

A video developed by researchers at RIKEN and the University of Tokyo using supercomputer-powered simulations of the human heart won the Best Visualization or Simulation award at the 42nd Computer Animation Festival in Los Angeles, United States. The event takes place every year during the SIGGRAPH conference on computer graphics and interactive techniques, and is recognized as a qualifying festival for the Academy of Motion Picture Arts and Sciences.

The award-winning video reveals detailed footage of the beating human heart generated using the multi-scale, multi-physics heart simulator UT-Heart and the power of the K computer that RIKEN operates. Based on knowledge gained from physics, engineering, medicine and physiology, UT-Heart models the behavior of thousands of contractile proteins to accurately calculate heart muscle and valve movement, blood flow, blood pressure and energy consumption. The video takes viewers on a journey into the heart, revealing the parachute-like structure of the mitral valve, and presents a fireworks display of how blood moves through the heart's four compartments.



The heart simulator UT-Heart and the K computer were used to generate detailed graphics of the beating human heart.

The researchers hope to use UT-Heart to develop therapies for cardiac diseases and to evaluate drugs. The video and related materials are available on the RIKEN HPCI Program for Computational Life Sciences website.
www.riken.jp/en/pr/topics/2015/20150618_1/

Stress-sensitive blood pressure regulation

A paper published in June 2015 in the journal *Molecular Cell* reports on a mechanism for regulating blood pressure uncovered by researchers at the RIKEN Brain Science Institute. The study could lead to new therapies for high blood pressure, which is a primary risk factor in stroke, heart disease and diabetes.

The RIKEN team noticed that mice lacking the protein ERp44 had about 20 per cent lower blood pressure than normal mice. The peptide hormone vital for maintaining blood pressure called angiotensin II was also found to go out of circulation in the ERp44-knockout mice faster than in normal mice. Further investigations revealed the underlying mechanism by which angiotensin II is removed from the blood to reduce blood pressure.

ERp44 is a multifunctional protein located in the endoplasmic reticulum, where proteins are folded into their proper shapes before being released into the cell. When oxygen levels in the endoplasmic reticulum are high, the enzyme ERAP1 binds to ERp44. But when oxygen levels are low, such as during cellular stress, ERAP1 is released into the bloodstream, where it cleaves angiotensin II. With no ERp44 to bind to in the knockout mice, ERAP1 was free to enter the bloodstream and digest angiotensin II, resulting in lower than normal blood pressure.
www.riken.jp/en/pr/press/2015/20150508_1/

The world's most powerful NMR

The most powerful nuclear magnetic resonance (NMR) system in the world, equipped with magnets that can generate a 1,020-megahertz magnetic field, has been developed by a team of researchers at the RIKEN Center for Life Science Technologies, Japan's National Institute for Materials Science, Kobe Steel, Ltd. and JEOL RESONANCE Inc., with support from the Japan Science and Technology Agency. The work took 20 years to complete, including recovering from damage caused by the 2011 Great East Japan Earthquake and a critical shortage in the global supply of helium, which is essential for cooling.

NMR is a technique used to determine the structural and chemical properties of compounds based on the nuclear spin of atoms under an external magnetic field—the stronger the magnetic field, the easier it is to detect these properties. The research team used high-temperature superconducting technology to produce the powerful magnetic field in their system, which will support drug development and research into structural biology, analytical chemistry and materials engineering. It could also translate into advances in similar technologies that rely on precise magnetic fields, such as magnetic resonance imaging.
www.nims.go.jp/eng/news/press/2015/07/201507010.html

Hideaki Maeda (second from right), director of the NMR Facility at the RIKEN Center for Life Science Technologies.

Tuberculosis's signature sugars

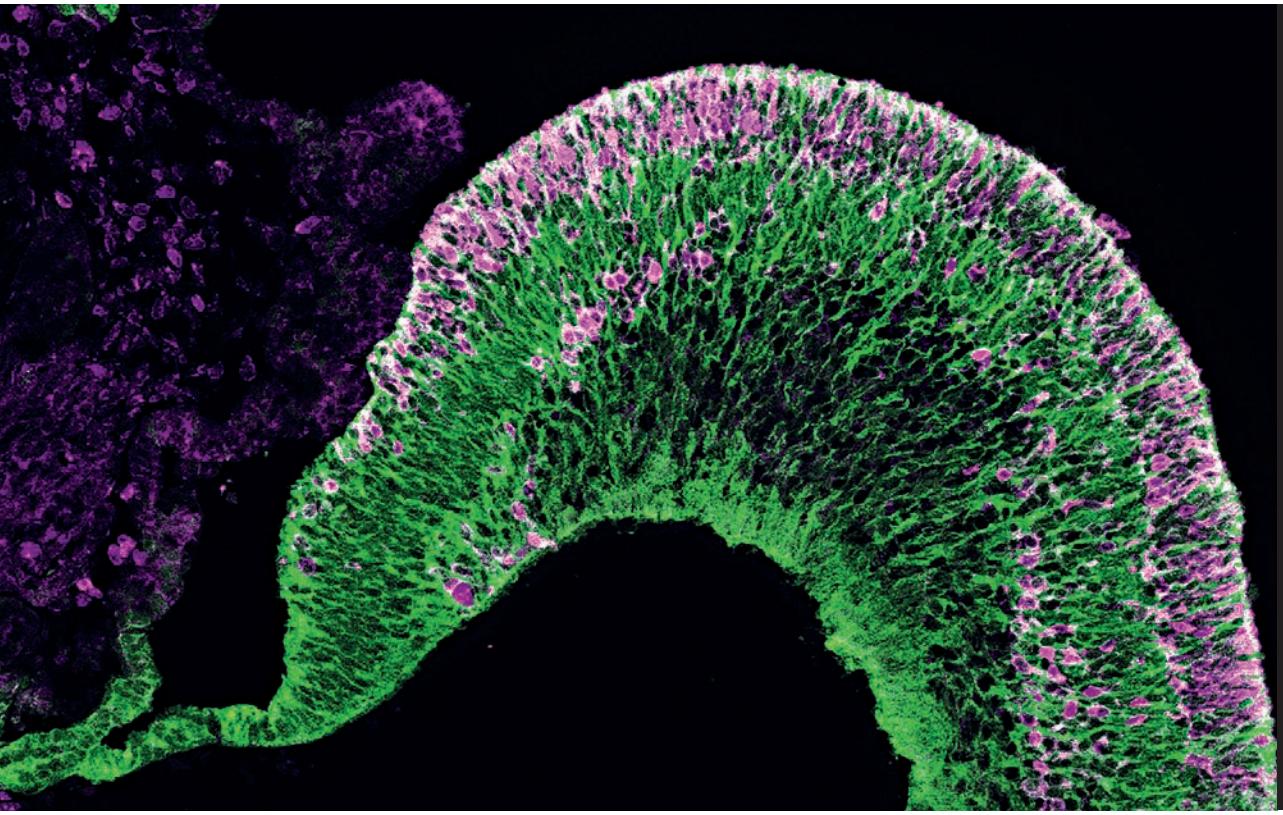
A protein produced in the pancreas can recognize the signature sugars attached to the tuberculosis-causing bacterium *Mycobacterium tuberculosis*, according to a study by an international team of researchers at RIKEN, the Max Planck Society and Imperial College London. "The study provides fundamental insights into how molecules in the tuberculosis bacteria are recognized by the human immune response," says Peter Seeberger, director of the Max Planck Institute of Colloids and Interfaces and a co-author of the study published in the journal *ChemBioChem* in July 2015.

The protein ZG16p helps to package enzymes in the pancreas and is known to be a lectin because it can bind to sugars in the cell membrane. Using a microarray that fixes many different sugars onto a microchip, the team found that ZG16p preferentially binds to two sugars that occur in *M. tuberculosis*—PIM1 and PIM2. They used atomic imaging techniques including nuclear magnetic resonance spectroscopy to identify hot spots in PIM1 and PIM2 that act as contact sites for ZG16p. Computer modeling provided additional insights into the binding characteristics of these complex molecules.

"Our finding is another example of the immense potential of the glycosciences," says Yoshiki Yamaguchi from the RIKEN–Max Planck Joint Research Center for Systems Chemical Biology, who led the study. "The PIM–lectin interaction mode revealed in this study may be useful for the development of therapeutic antibodies or vaccines."

www.riken.jp/en/pr/topics/2015/20150522_2/

Research highlights



Fluorescence microscopy image showing the self-organization of discrete populations of different eye cells formed from embryonic stem cells to construct part of the neural retina.

BIOLOGY

Growing eyes from stem cells

Embryonic stem cells give rise to three-dimensional, multilayered retinal tissue in a dish

A RIKEN team has succeeded in developing a culture method that allows human embryonic stem cells to be efficiently and spontaneously grown into the many cell types found in the human retina[1]. The achievement brings scientists a step closer to growing the most complex component of the eye—the eye's neural tissue—and could enable doctors to repair damaged eyes with lab-grown retinal tissue.

Yoshiki Sasai, Mototsugu Eiraku and their colleagues from the RIKEN Center for Developmental Biology had previously shown that they could coax human and mouse embryonic stem cells to form retinal progenitors. After a few weeks in culture, these cells would spontaneously self-assemble into three-dimensional retinal tissue. Yet although the structure contained multiple layers of neural retinal cells, including both rod and cone photoreceptors, the long-term organization of these cells was not entirely clear.

The RIKEN team, collaborating with Sumitomo Chemical Company, refined and improved this culture technique through precisely timed treatment with a regulatory protein called BMP4. This improved method transforms human embryonic stem cells into retinal progenitors without the addition of extracellular matrix products required previously to promote retinal differentiation. This allows for a more controlled process that could be suitable for future clinical applications.

The researchers also developed a method to create the ciliary margin, which resides at the boundary between the neural retina and the retinal pigment epithelium. Their step-wise 'induction-reversal' culture method induces the formation of boundary tissue, which then self-organizes into ciliary margin tissue. Retinal stem cells residing in the ciliary margin generate new retinal tissue and help contribute to retinal tissue growth in the culture dish. This degree of cellular organization (see image) is the closest scientists have yet come to building self-growing retinal tissue from stem cells.

"Our results are consistent with the current view that the retinal pigment epithelium and neural retina are capable of 'fate transition,'" says Eiraku. "By examining retinal formation in culture, we hope to reveal the mechanisms involved in human retinal development."

The methods developed by Eiraku and his colleagues could one day be used to culture tissue that can be transplanted into a human retina damaged by conditions such as macular degeneration and retinitis pigmentosa, which lead to blindness. "The protocol developed here allows us to generate retinal tissue that closely resembles the biological retina with high efficiency and stability," notes lead author Atsushi Kuwahara. "It is a step closer to realizing regenerative medicine for retinal disorders." ■

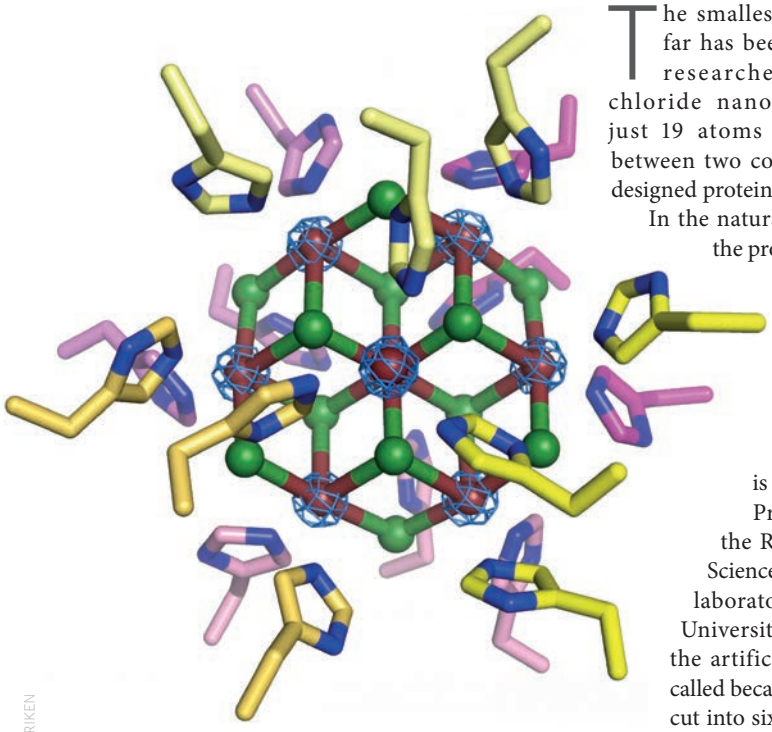
Reference

1. Kuwahara, A., Ozone, C., Nakano, T., Saito, K., Eiraku, M. & Sasai, Y. Generation of a ciliary margin-like stem cell niche from self-organizing human retinal tissue. *Nature Communications* **6**, 6286 (2015).

CHEMISTRY | PRESS RELEASE

Waiter, there's a nanocrystal in my pizza!

A tiny crystal between two artificially engineered proteins could provide valuable insights into how proteins in nature incorporate metals into their structures



The structure of a cadmium chloride nanocrystal, the smallest crystal reported so far.

The smallest crystal reported so far has been created by RIKEN researchers. The cadmium chloride nanocrystal consists of just 19 atoms and is sandwiched between two copies of an artificially designed protein[1].

In the natural world, proteins use the process of biomineralization to incorporate metallic elements into tissues, creating diverse materials such as seashells, teeth and bones. However, this process is not well understood.

Previously, scientists at the RIKEN Center for Life Science Technologies and collaborators at Yokohama City University in Japan developed the artificial protein Pizza6, so called because it resembles a pizza cut into six identical slices. Their goal was to design novel proteins that do not exist in nature and that could be used in various applications. Proteins like Pizza with its high degree of symmetry are attractive scaffolds for creating new hybrid biomaterials suitable for applications such as drug packaging and delivery to cells, or even bioremediation of hazardous metals in the environment.

In the current study, the researchers modified the Pizza protein by introducing a metal-binding site. "Our initial impetus was to design metal-binding sites to control the self-assembly of our designed symmetric proteins," says first author Arnout Voet. "We used computational methods to find a rational way to incorporate a metal-binding site into the Pizza protein we had previously designed, based on the idea that this could allow us to control protein assembly easily." The scientists anticipated that this would provide a new tool for building novel proteins from the ground up by using very cheap metal reagents.

When the proteins were modified to have a metal-binding site and then placed in a solution of cadmium chloride, multiple subunits spontaneously bound together to form one copy of Pizza. Using RIKEN's SPring-8 synchrotron and other facilities, the researchers analyzed the

structure at the atomic level and discovered that the atoms of cadmium and chloride had formed a tiny lattice—a crystal structure—sandwiched between two Pizza proteins (see image).

"We were very excited to see the formation of the crystal, as it provides insights into the process of biomineralization," says Kam Zhang, who led the RIKEN team. "Our results indicate the feasibility of using rationally designed symmetric proteins to biomineralize nanocrystals. Achieving this could allow us to make a wide range of nanodevices such as biopharmaceuticals, biosensors, light-driven switches and synthetic enzymes from the bottom up."

"We have many ideas about how this might be put to further use," Zhang continues, "and will continue to experiment to find novel properties in these artificially designed proteins." ■

Reference

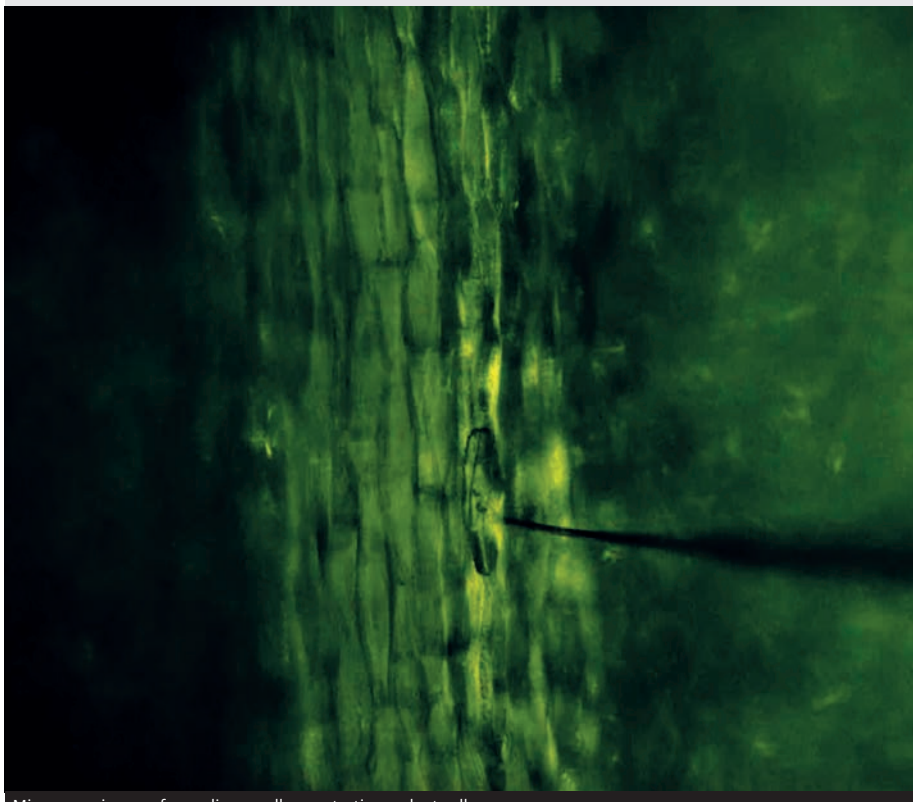
1. Voet, A. R. D., Noguchi, H., Addy, C., Zhang, K. Y. J. & Tame, J. R. H. Biomineralization of a cadmium chloride nanocrystal by a designed symmetrical protein. *Angewandte Chemie International Edition* **54,** 9857–9860 (2015).

BIOLOGY

A stress tracker for plants

Measuring stress-related hormones in individual plant cells reveals local patterns of metabolic activity

A new technique that allows stress responses in plant leaves to be monitored using single living cells could allow plant breeders to rapidly and non-destructively measure hormones, nutrient levels, pesticide residues or any cellular compound of interest[1]. The method, developed by a team led by Mitsunori Seo from the RIKEN Center for Sustainable Resource Science, is expected to be particularly useful for quality testing of agricultural products.



Microscopy image of sampling needle penetrating a plant cell.

Plants respond to stresses by releasing minute quantities of hormones in a precise, localized fashion. For example, hormones known as jasmonates accumulate at the sites of cuts to induce wound healing, while during times of water scarcity, levels of another hormone called abscisic acid increase at microscopic pores on the surface of leaves to induce the pores to close.

Standard methods for measuring plant hormones require large amounts of plant tissue. "But hormone levels determined from a mixture of cells do not always tell us what is actually occurring within plants," notes Seo.

To better understand the local distribution of hormones, Seo and his colleagues developed a method for quantifying hormone levels in individual cells. Working with the broad bean *Vicia faba*, the research team, which also included scientists from the RIKEN Quantitative Biology Center, applied a technique called nanoelectrospray ionization to add an electric charge to molecules inside single plant cells.

Obtaining samples from single cells was challenging. "Plant cells are surrounded by thick cell walls," says Seo. "It took a lot of practice, but eventually we developed a reliable technique." With the samples in hand, the researchers then measured the abundance of hormones using a common analytical chemistry instrument called a mass spectrometer.

In this way, the researchers detected abscisic acid in individual water-stressed cells and jasmonoyl-isoleucine—a type of

jasmonate—in single wound-stressed cells. In contrast, both hormones were largely undetectable in broad bean leaf cells grown under non-stressed conditions.

Tracking abscisic acid and jasmonate levels could be just the tip of the analytic iceberg. According to Seo, the same basic methodology should be widely applicable beyond these two hormones. "Our method can be used to determine the levels of other hormones as well as primary and secondary metabolites," he says. "However, we must optimize the procedures for each compound."

The researchers continue to refine and improve the technique and also plan to develop quality control measures to account for potential impurities and for the variability in volume between different cells. ■

Reference

1. Shimizu, T., Miyakawa, S., Esaki, T., Mizuno, H., Masujima, T., Koshiba, T. & Seo, M. Live single-cell plant hormone analysis by video-mass spectrometry. *Plant & Cell Physiology* **56,** 1287–1296 (2015).

PHYSICS | PRESS RELEASE

Stable magnetic vortices observed at room temperature

Materials that host stable magnetic vortices at room temperature are promising for the development of high-density memories and other devices

A class of materials that can host stable magnetic vortices known as skyrmions at room temperature and above has been discovered by an international team of researchers, paving the way for the development of useful 'spintronic' devices[1].

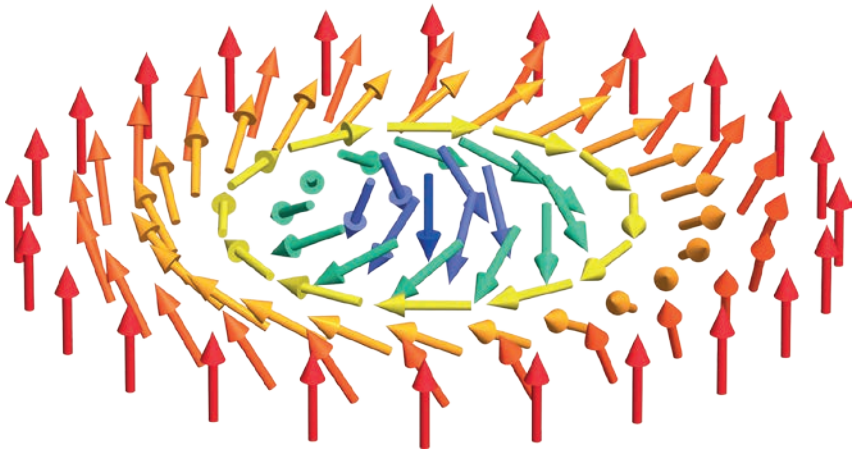
Magnetic skyrmions (see image) are tiny, nanometer-sized vortices of magnetic spin that emerge in the bulk of magnetic materials. Their small size could enable them to be used in extremely dense memory devices, which use the presence or absence of skyrmions as bits in computer calculations. This could lead to high-density magnetic memories that have ultralow-power consumptions.

However, skyrmions are not always easy to use. Although they occur ubiquitously in magnets under a wide range of conditions, they can be tricky to control. The ultimate dream of researchers has been to create stable skyrmions that have a set chirality and do not decay.

Skyrmions emerging from a process called the Dzyaloshinskii–Moriya interaction are considered to be particularly promising, as they can be small (under 150 nanometers) and have a fixed spin direction. Consequently, materials in which this interaction occurs could host a large number of stable skyrmions, which could be manipulated by electric currents, and hence be used to create high-density data-storage devices. Such stable skyrmions have been found in certain crystal structures, but the skyrmions emerge only at low temperatures in the materials studied to date.

In the present study, Yusuke Tokunaga, Yasujiro Taguchi and their colleagues from the RIKEN Center for Emergent Matter Science, along with collaborators in Europe, examined a magnetic material consisting of cobalt, zinc and manganese whose structure seemed likely to host stable skyrmions. Using various techniques, they found that when a magnetic field was applied to either a block or a thin plate of the material, it clearly exhibited skyrmion crystals. These skyrmions were both stable and chiral, so that they spun in a set direction.

"We are quite excited about these results, as they may answer the long-held expectation that we can find skyrmion-hosting



A schematic representation of a magnetic skyrmion, where the arrows represent the spin direction.

systems in a variety of new materials," says Tokunaga. "In addition, the fact that we have shown that skyrmions can be stabilized at room temperature and above opens the road to looking for ways to integrate skyrmions into spintronic devices without complicated cooling systems." ■

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BIOLOGY

Uncovering the secrets of antibacterial response

New insights into a well-known immune pathway could help researchers fight tuberculosis and other infectious diseases

A protein with a previously unrecognized—but apparently critical—role in a well-studied immune pathway has been identified by a team jointly led by Harukazu Suzuki of the RIKEN Center for Life Science Technologies in Japan and Frank Brombacher in South Africa[1].

Immune cells known as macrophages fend off pathogenic bacteria by destroying infected cells. This process triggers a signaling cascade known as the classical activation pathway, which in turn stimulates a broader inflammatory response. However, despite extensive study, many of the molecular mechanisms that govern the complex transcriptional regulation in classically activated macrophages are not well understood.

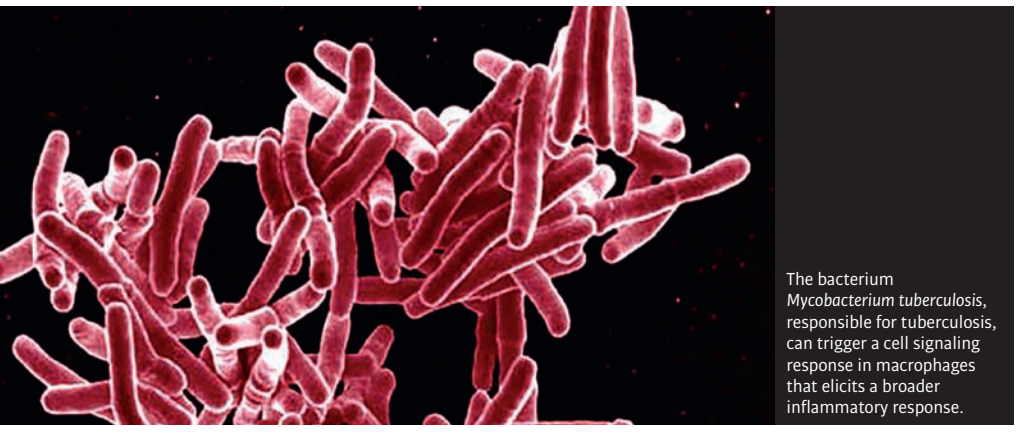
Since the protein interferon-γ (IFN-γ) helps initiate classical activation, the researchers initially set out to identify genes switched on by IFN-γ. They observed particularly strong activity from *Batf2*, which expresses a protein belonging to a family of transcription factors that regulate genes but are otherwise poorly characterized. The scientists found that they could interfere with many aspects of IFN-γ-induced classical activation by experimentally manipulating macrophages to reduce *Batf2* activity.

The bacterium responsible for tuberculosis, *Mycobacterium tuberculosis* (see image), can also trigger classical activation, and the researchers observed an equally prominent role for *Batf2* in this response. Indeed, many genes activated by the Batf2 protein make important contributions to host defense and to promoting inflammation, providing further evidence that the gene is a valuable mediator of classical activation.

Although the Batf2 protein can switch genes on and off, it lacks the structural components necessary to bind to DNA. Consequently, it has to rely on a DNA-binding 'partner'. By examining the sequences of known Batf2 target genes, Suzuki and his co-workers zeroed in on IFN regulatory factor-1 (Irf1) as the most likely collaborator. 'Knockdown' experiments that reduced macrophage production of Irf1 confirmed this, with many of the same changes in gene expression observed as in the *Batf2*-deficient cells.

This discovery of a major component of the macrophage response that had gone unnoticed for so long surprised Suzuki. "This response has been extensively analyzed by many groups," he says. "I initially thought that Batf2 might be a minor modulator of classical activation, but I now believe that Batf2 plays a central role, together with Irf1."

Suzuki's group intends to delve deeper into the nuts and bolts of the machinery for macrophage activation. Suzuki is also working with Brombacher to learn whether this pathway might offer a target for stimulating the immune response against tuberculosis. "We are currently analyzing the *in vivo* effect of Batf2 on *M. tuberculosis* infection in mice," he says. ■



The bacterium *Mycobacterium tuberculosis*, responsible for tuberculosis, can trigger a cell signaling response in macrophages that elicits a broader inflammatory response.

Courtesy: National Institute of Allergy and Infectious Diseases

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BIOLOGY | PRESS RELEASE

Summer on the brain

Brain activity involving a neural transmitter and chloride levels allows mice to track the seasons

A key mechanism enabling animals to keep track of the seasons has been uncovered by researchers led by Toru Takumi at the RIKEN Brain Science Institute. They show how the brain's circadian clock machinery encodes seasonal changes in the duration of daylight through the activity of the neurotransmitter GABA along with changes in chloride levels in certain neurons[1].

Seasonal time keeping is important for both animals and people. It is accomplished by the same part of the brain that governs daily circadian rhythms—the suprachiasmatic nucleus (SCN). The SCN cyclically expresses certain 'clock' genes during a 24-hour period. But not all of the neurons march to the same beat—two regions in the SCN are slightly out of phase; as day length increases, so does the phase gap between them.

To understand how this happens, the researchers measured expression levels of the clock gene *Bmal1* in explanted dorsal and ventral SCNs of mice that had been living in long- or short-day light cycles. As expected, cyclical *Bmal1* levels in dorsal and ventral regions from the long-day group were out of phase, whereas those from the short-day group were synchronized. Modeling analysis predicted that the asymmetry between the two regions causes the dorsal region to become out of phase when daylight increases.

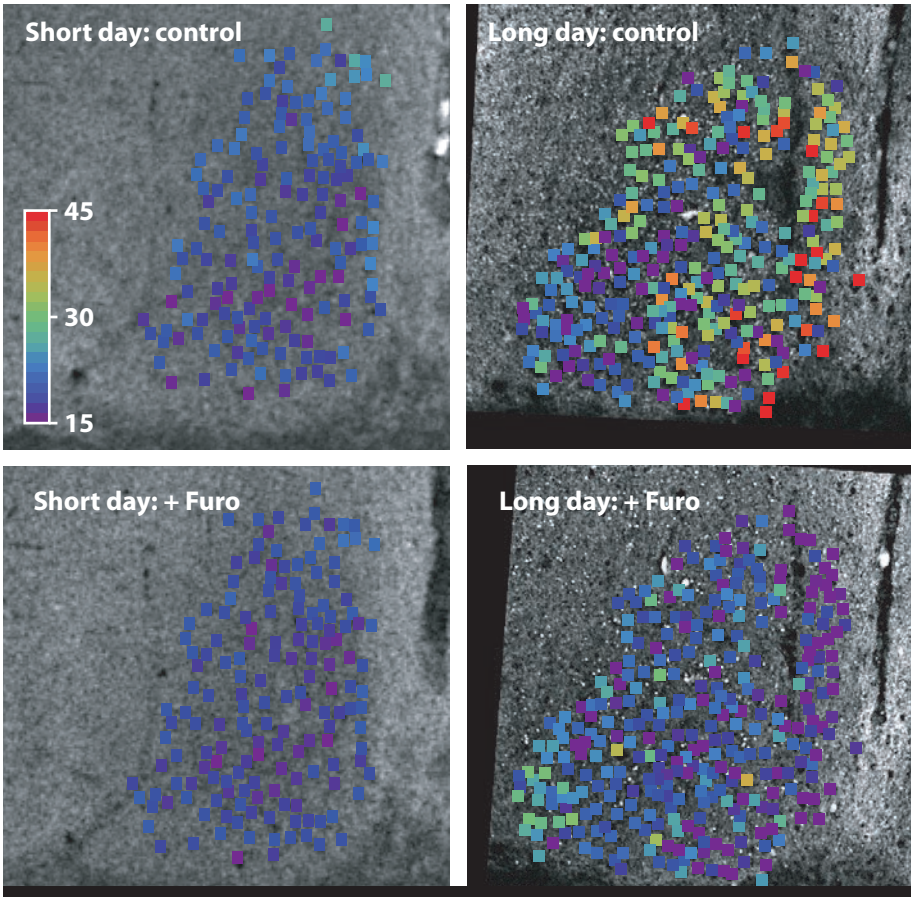
The researchers found that GABA generally inhibits neuronal activity but that some SCN neurons are actually excited by GABA.

"GABA becomes excitatory when chloride levels inside neurons are high," explains lead researcher Jihwan Myung. "We suspected that changes in GABA function across the SCN could represent the repulsive force that pushes these two clusters of neurons out of phase."

When the researchers blocked GABA activity, the large phase gap seen in the long-day group disappeared and the cycles of *Bmal1* levels came to resemble those of the short-day group. The researchers conjectured that GABA has a special effect on the dorsal SCN.

To test this, they measured expression levels of two genes responsible for importing and exporting chloride. The team found that in long-day SCNs, the expression ratio of the two genes in the dorsal SCN changed, resulting in significantly more chloride being imported and making the effect of GABA preferentially excitatory in the dorsal region; blocking chloride import removed the phase gap in the long-day group.

"Sudden changes in seasonal day length can cause severe mood disorder in some individuals." says Myung. "Understanding how to adjust our internal seasonal clock could lead to effective ways of helping people whose internal clocks have been disrupted." ■



Compared to exposure to short days (top left), exposure to long days leads to increased chloride in the suprachiasmatic nucleus (SCN) (top right). Inhibiting chloride import with furosemide (Furo) reduces chloride levels only in SCNs exposed to long days (bottom right). Color bar indicates intracellular chloride concentration in micromoles per liter.

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Biology

The power of positive memories

Stress-induced depression can be overcome in mice by inducing the firing of neurons that had been active during a positive experience

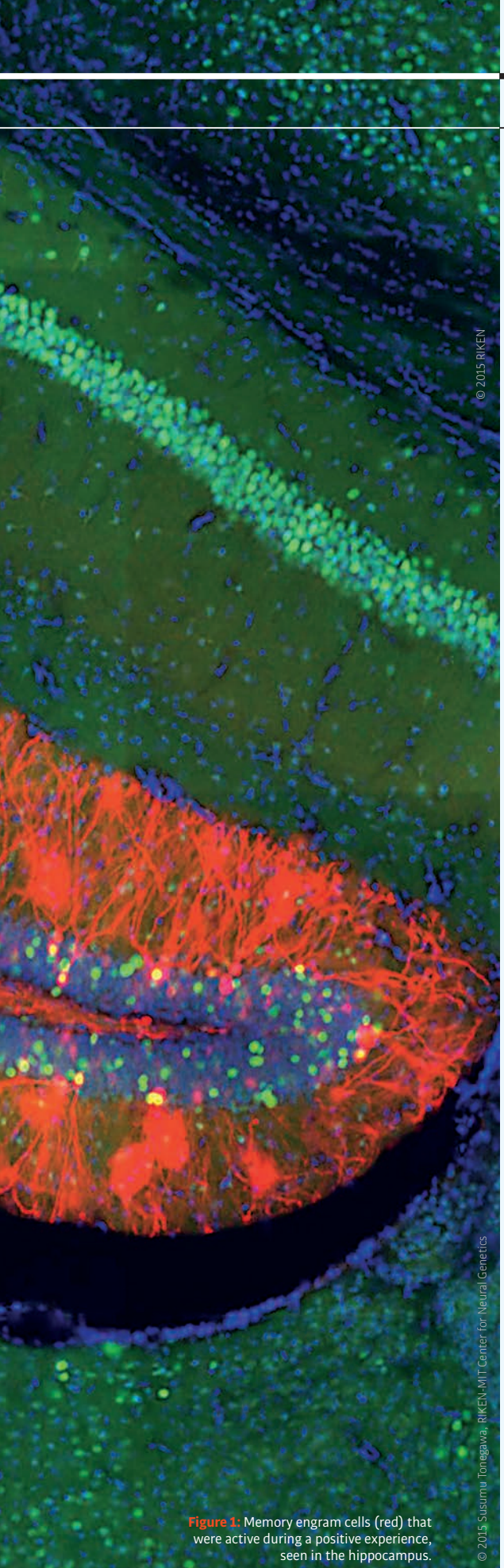

Figure 1: Memory engram cells (red) that were active during a positive experience, seen in the hippocampus.

Chronic stress can cause symptoms of anxiety and depression in many organisms, including mice and humans. A team of RIKEN researchers has discovered that stimulating brain cells that had previously fired during a positive experience blocks the stress-induced onset of depressive behaviors in mice[1]. This finding may assist in identifying the circuits that need to be targeted to treat depression in people.

Susumu Tonegawa and his colleagues at the RIKEN–MIT Center for Neural Circuit Genetics at the Massachusetts Institute of Technology (MIT) exposed one population of male mice to the rewarding experience of spending time with a female, and another population to the stressful experience of being immobilized. All the mice had been genetically engineered so that only neurons activated during these two experiences would express a special protein channel, which the researchers could later artificially stimulate by shining a blue light into the brain.

Previous studies had shown that mice exposed to chronic stress exhibit depression-like behaviors, such as loss of interest in drinking a sugary solution and a lack of effort to right themselves when suspended by their tails. Stressed mice also demonstrate greater anxiety than unstressed mice. For example, stressed mice tend to stick to the edges of an open field instead of venturing out to explore the more exposed center.

In their latest experiment, Tonegawa and his colleagues discovered that reactivating neurons that had previously been active during a positive experience (Fig. 1) prevented depression-like behaviors in mice exposed to chronic stress. But they found that the same effect did not occur when they reactivated neurons that had been activated during a negative experience (Fig. 2).

However, stimulating these neurons did not block the induction of anxiety. These findings suggest that other parts of the brain not targeted by the reactivation approaches in this study may modulate stress-induced anxiety.

Making new neurons

A structure in the brain known as the hippocampus exhibits many changes in response to stress and depression. For example, both stress and depression have been shown to reduce new neuron formation, or neurogenesis, in the hippocampus. Tonegawa and his colleagues found that mice exposed to chronic stress had fewer newly generated neurons than mice that had not been stressed. But they also discovered that light-induced reactivation of neurons that had been active during a positive experience restored neurogenesis in the hippocampus of stressed mice.

Antidepressants, which are used by millions of people worldwide, can induce hippocampal neurogenesis if taken for extended periods of time. In contrast, Tonegawa's team observed increased

Susumu Tonegawa

Susumu Tonegawa received his doctorate from the University of California, San Diego, in the United States. He then undertook postdoctoral work at the neighboring Salk Institute for Biological Studies before moving to the Basel Institute for Immunology in Switzerland, where he performed his landmark immunology experiments. In 1987, Tonegawa was awarded the Nobel Prize in Physiology or Medicine. Using advanced genetic manipulation techniques, Tonegawa is now unraveling the molecular, cellular and neural circuit mechanisms that underlie learning and memory. He is currently director of both the RIKEN–MIT Center for Neural Circuit Genetics at the Massachusetts Institute of Technology and the RIKEN Brain Science Institute in Japan.

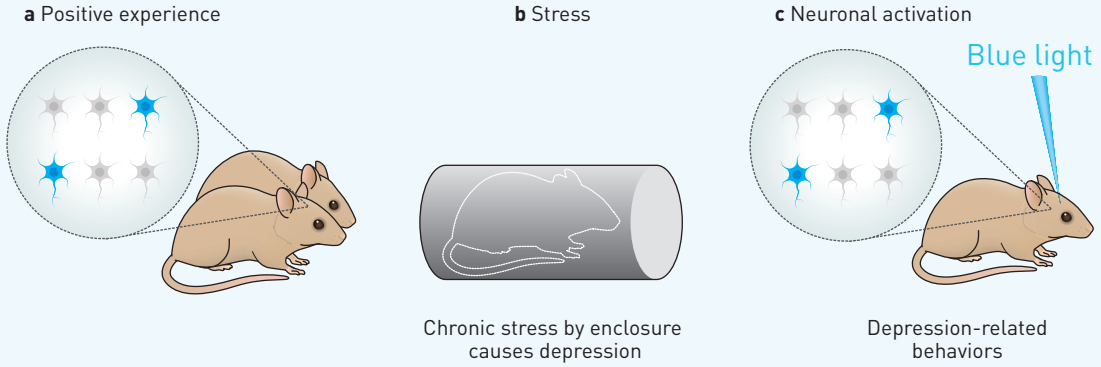

Figure 2: By using light to reactivate neurons that were active during a positive experience, researchers have found a way to reverse depression-related behaviors in mice. The same effect was not observed for mice subjected to the stressful experience of being immobilized.

neurogenesis after just five days of reactivating neurons that had been active during a positive experience. This enhanced neurogenesis may be tied to the beneficial effects they observed in their mice. The researchers suggest that the brain plasticity—a concept used to describe the brain's ability to change in response to new stimuli and experiences—that takes a long time for antidepressants to achieve could be rapidly mobilized by identifying and stimulating neurons linked to a positive experience.

Pinpointing critical brain cell circuits

Many parts of the brain may work together in a circuit to encode positive experiences and their influence on behavior. Tonegawa and his co-workers used light to stimulate the hippocampal neurons that had previously been labeled during exposure to a positive experience. They then used a marker to label neurons in other brain areas that were activated by the hippocampal cells, including the basolateral amygdala and the nucleus accumbens, two brain areas known to play a role in modulating behavioral responses to reward.

When the researchers used gene-therapy approaches to prevent neurons in the basolateral amygdala from firing, the stimulation of hippocampal neurons that encode positive experiences ceased to induce antidepressant responses in stressed mice. This suggests that the circuit from the hippocampus to the basolateral amygdala plays a key role in encoding how positive experiences can block the effects of chronic stress.

Many axons that utilize the neurotransmitters glutamate and dopamine extend from the basolateral amygdala to the nucleus accumbens, and the connections between these brain structures are known to be involved in responses to stressful events. The researchers showed that administering drugs in the nucleus accumbens that block dopamine or glutamate suppresses the antidepressant-like effects of light stimulation of hippocampal cells linked to a positive experience. Therefore, they conjecture that this circuit may be a nexus in which information linking a stimulus with a reward combines with the current motivational landscape in which an animal finds itself to drive behavioral outcomes.

Fighting depression

The findings by Tonegawa and his colleagues may shed light on a neurosurgical technique for treating depression known as deep brain stimulation. This technique involves implanting a stimulator within the brain cortex of depressed individuals who have failed to respond to more conventional pharmacological treatments. The extent to which the circuits and pathways identified in this study play a role in the beneficial effects of deep brain stimulation remains to be explored. If these same circuits and neurotransmitter systems are linked to the encoding or the recall of positive experiences in humans during stressful life events, the findings could lead to new therapeutic approaches for fighting depression in people.

The research points the way forward to a radically new approach for treating depression. "Most depression therapies focus on systemic drugs," notes Tonegawa. "Our findings suggest that treatment may be achievable by stimulating specific neural circuits."

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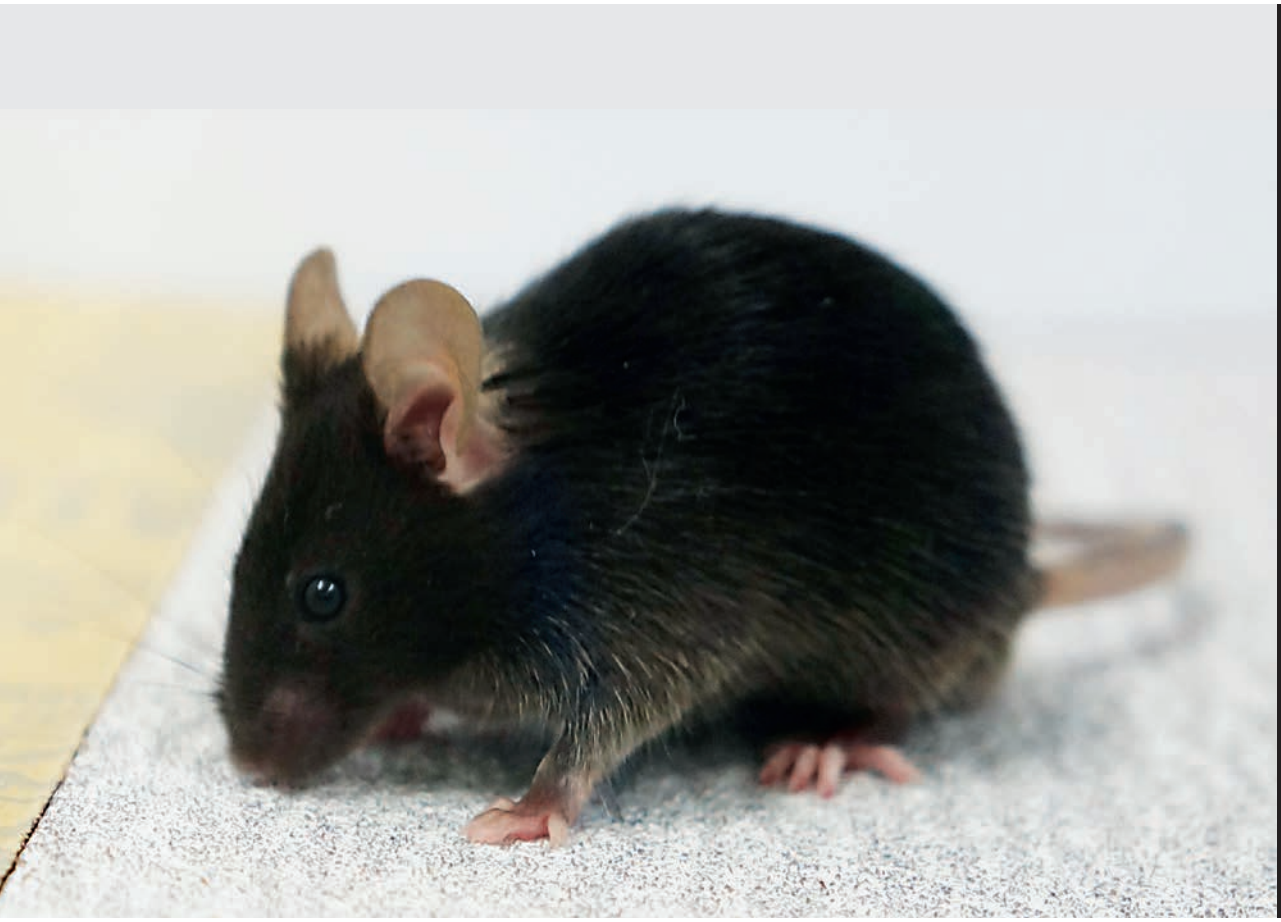

A mouse uses tactile sensation in its footpads to detect the rough texture of the floor.

BIOLOGY | PRESS RELEASE

Taking the rough with the smooth

Disabling a circuit in the brain renders mice unable to distinguish between different textures

The sense of touch is vital, but we often take it for granted because it seems simple and automatic. A RIKEN study suggests that this apparent simplicity comes from a clever two-stage brain circuit. When researchers manipulated this circuit with light-driven optical genetic tools, laboratory mice (see image) became unable to distinguish between rough and smooth surfaces[1].

A team at the RIKEN Brain Science Institute showed that the perception of touch relies on two signals, one from the skin to the brain and another within the brain itself. This second signal relays the first signal from a lower level brain area to a higher one and then boomerangs it back to the lower level.

When the researchers, led by Masanori Murayama, observed the brains of mice after touching their paws, they saw immediate activity in the sensory cortex—the brain area that receives signals from the skin. Unexpectedly, they also recorded a second source of activity tens of milliseconds after the first.

"We investigated the source of this second activation and found that high-level motor cortex receives information from the sensory cortex and sends it back to the sensory cortex." Murayama explains. "This means that, for tactile perception, the flow of information from the skin to brain requires communication not only from the periphery to the brain but also reverberation between two brain areas."

It was previously thought that one signal from the skin to the brain was sufficient to produce touch sensation, but this study

revealed that without the second signal, mice cannot feel or use the incoming sensory information, suggesting that they may not even perceive texture differences.

To investigate this idea, the researchers trained mice to distinguish between rough and smooth floor textures by associating one of them with a food reward. When they disabled the second signal by shutting off the responsible neurons with light-activated optical genetic technology, the mice could not distinguish the two floor textures.

"Our results show that a reverberant neural circuit from the sensory cortex to higher motor cortex is required for the perception of touch," says lead researcher Satoshi Manita. Murayama speculates that this two-stage circuit design may be a safety mechanism to ensure continuous, accurate perception even when distracted by other senses. "This form of perception, an external signal and its internal rebound, may extend to other senses," he concludes, "and we may find that communication between brain areas refines perception for more accurate and integrated behavior." ■

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BIOLOGY

The chick egg gives up its secrets

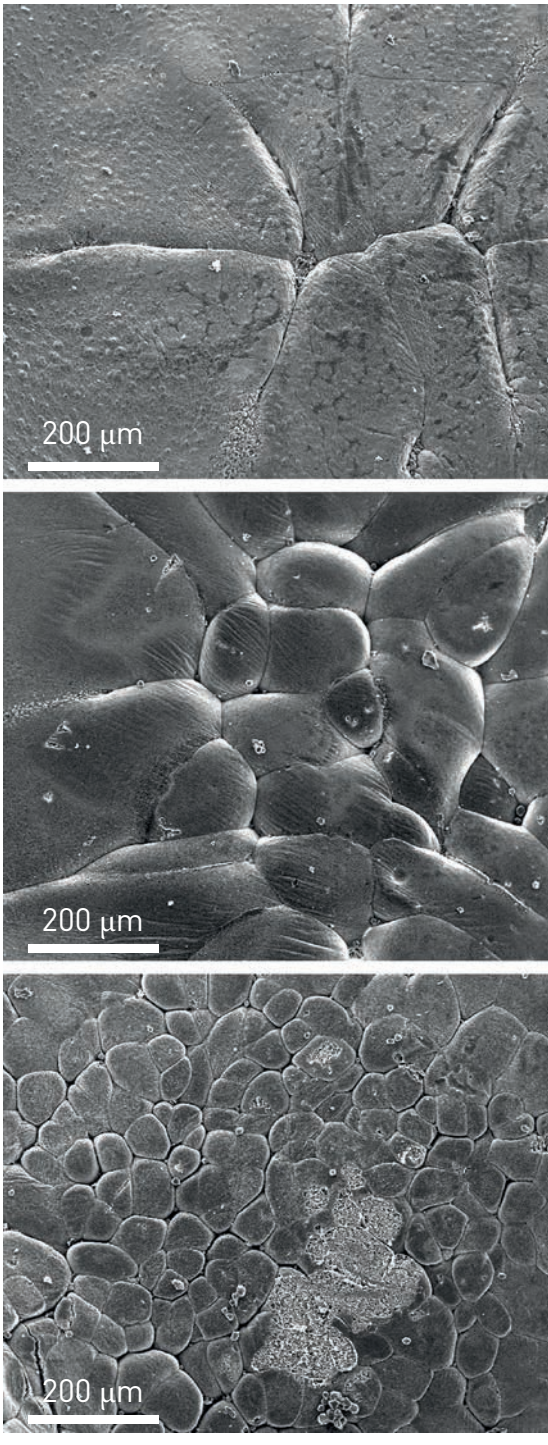
Analysis of early chick embryos provides unique insights into early vertebrate development and evolution

The mysterious period of early chick development that occurs inside the mother hen has been revealed for the first time by a collaborative team involving researchers from RIKEN, Seoul National University in Korea and Shizuoka University in Japan[1]. The study demonstrates that despite apparent differences, vertebrate embryos undertake very similar cellular journeys and share fundamentally conserved developmental mechanisms.

Early chick development was generally characterized in the 1970s, but no further cellular or molecular details of the crucial first 25 hours post-fertilization have been subsequently reported. As Guojun Sheng from the RIKEN Center for Developmental Biology explains: "The chick is notorious as the only model animal commonly used in developmental biology whose early phase of development is inaccessible for experimental analysis."

Within 13 hours of fertilization, the fertilized chick egg develops through a rapid series of cell cycles to produce cell clusters. This is known as the 'cleavage' stage of embryonic development; molecular and cellular events taking place during this stage are crucial for later lineage specification. Little is known, however, about these events in chick embryos. For example, like other vertebrates that develop from a yolk-rich egg, chick embryos begin to direct their own development relatively early, but the exact timing of this transition, known as zygotic gene activation, is unknown.

To investigate, Sheng and his colleagues collected early cleavage stage chick embryos from laying hens using a non-invasive abdominal massage method and examined the embryos by scanning electron microscopy. The research team was able to reconstruct the development of the embryo over the course of a few hours, from 8 open cells nestled in furrows in the yolk surface,



Scanning electron microscopy images show the development of the chick embryo from eight open cells in yolk furrows (top) to laterally closed cell clusters (bottom).

to 16 closed cells that then kept multiplying to form a four-cell-thick layer of cell clusters (see image). They also observed the formation of a cavity above the yolk surface and a yolk syncytial layer similar to that observed in fish embryos.

Using a marker of gene transcription, Sheng and his colleagues were able to determine that zygotic gene activation takes place after about eight cell cycles in chick embryos. In mammals, zygotic gene activation takes place before the third cell cycle, whereas in fish and frog embryos, it does not occur until the eighth or ninth cell cycle. The results suggest the existence of an evolutionary conserved mechanism for control of zygotic gene activation in yolk-rich embryos. "My aim is to convince people that the chick is a great model for understanding early human development," says Sheng. ■

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PHYSICS | PRESS RELEASE

Light shares spin effect with electrons

Although photons behave very differently from electrons, they exhibit a quantum spin phenomenon that had until now only been observed for charged particles

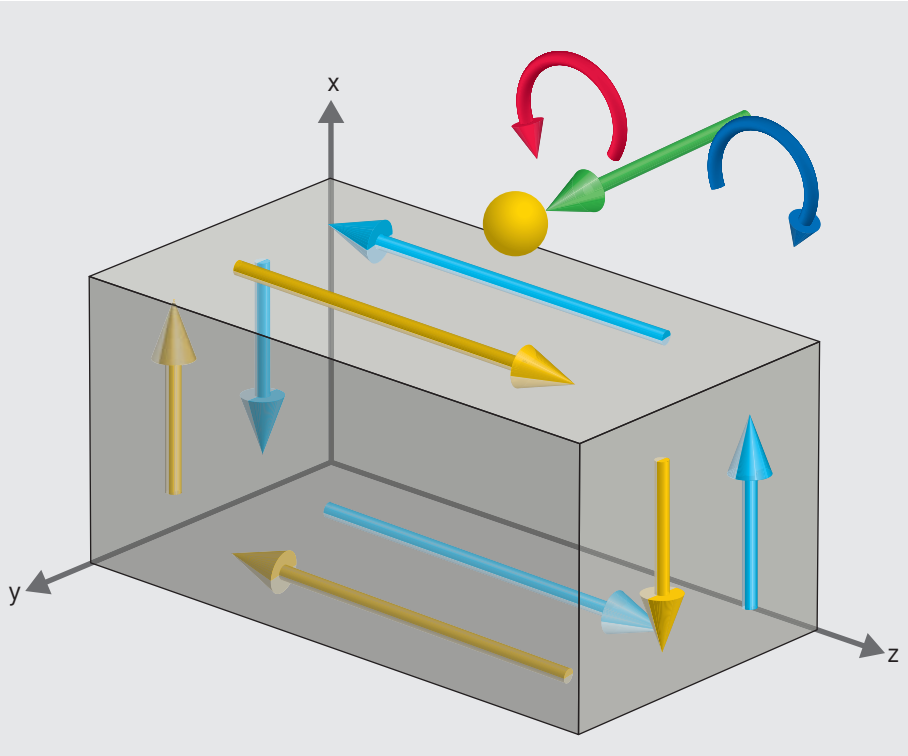
A solid-state physics phenomenon known as the quantum spin Hall effect is also an intrinsic property of light, according to a team of RIKEN researchers led by Konstantin Bliokh of the RIKEN Center for Emergent Matter Science[1].

Photons have neither mass nor charge, and so behave very differently from massive particles. But they do share a property called spin—a measure of the intrinsic angular momentum. Quantum spin can be thought of as an equivalent to the spin of a top, and it results in remarkable geometric and topological phenomena. The researchers discovered that, like electrons, photons exhibit a property related to spin—the quantum spin Hall effect.

The researchers had previously explored electromagnetic waves known as evanescent waves, which propagate along metal surfaces, for example, in much the same way that ocean waves emerge at the interface between air and water.

Bliokh says: "We realized the remarkable property we found, an unusual transverse spin, was a manifestation of the fact that free-space light exhibits an intrinsic quantum spin Hall effect, meaning that evanescent waves with opposite spins will travel in opposite directions along an interface between two media."

The quantum spin Hall effect for electrons allows for the existence of an unusual class



Schematic of experiments demonstrating the quantum spin Hall effect for light. Light propagating in the y direction (shown in green) is coupled to surface evanescent modes (shown in yellow and light blue). Depending on the spin of the incident light (shown by the red and dark blue circular arrows), surface waves are excited with opposite directions of propagation.

of materials called topological insulators that conduct electricity on their surfaces but not in their interiors. The team was intrigued to learn that analogues of these materials exist for photons. Although light does not propagate through metals, it can propagate along interfaces between a metal and air, in the form of so-called surface plasmons involving evanescent light waves. The group showed that the unusual transverse spin they found in evanescent waves can be associated with the intrinsic quantum Hall effect of photons (see image). Their findings explain recent experiments that have shown spin-controlled propagation of surface optical modes in one direction.

"On a purely scientific level, this research deepens our understanding of the classical theory of light waves developed by James Clark Maxwell 150 years ago," says Bliokh. "And it could also lead to applications using optical devices that are based on the direction of spin."

Franco Nori, who organized the project, says, "This work was made possible by the interdisciplinary nature of RIKEN, as we were able to bring together discoveries made in several different areas, to show that transverse spin, locked to the direction of propagation of waves, seems to be a universal feature of surface waves, even when they are of a different nature." ■

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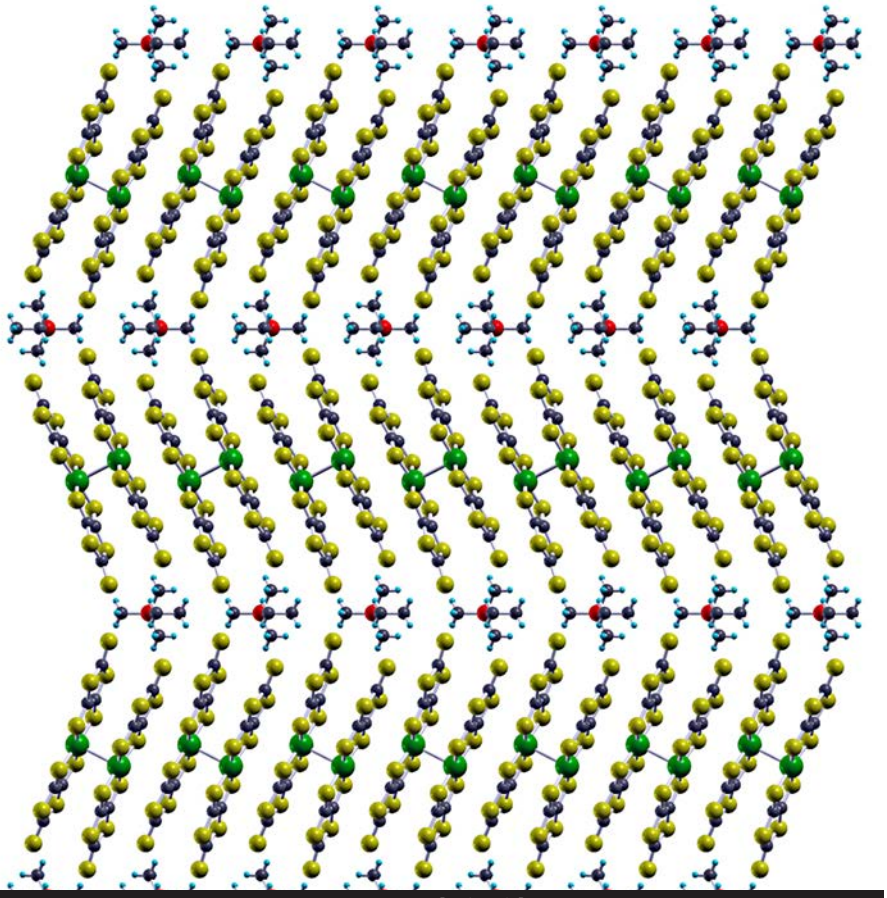
CHEMISTRY

A universal transition

Organic molecules reveal a universal behavior that governs the transition of many materials from an insulator to a conductor



The layered crystal structure of the organic conductor $EtMe_3P[Pd(dmit)_2]_2$.

Understanding what causes materials to change from electrical insulators to metallic conductors is relevant not only to the development of practical electronic devices, but also for fundamental insight into the physical properties of materials. The organic material $EtMe_3P[Pd(dmit)_2]_2$ is an insulator that becomes a conductor under certain conditions. It also has a number of unusual properties owing to the relationship between some of its energy states and its crystal structure.

By studying this transition in detail, Majed Abdel-Jawad from the RIKEN Condensed Molecular Materials Laboratory and co-workers have now discovered that the insulator-to-metal transition shows a universal behavior that applies to all related materials[1]. "This means that once a phase transition is associated with such a universality class, all physical properties of this transition can be predicted," explains Abdel-Jawad.

$EtMe_3P[Pd(dmit)_2]_2$ is an organic charge-transfer salt in which half of the electronic states that can contribute to the material's electrical conductivity are occupied by electrons, and the other half are empty. Usually, this would mean that the material is a good metallic conductor, because electrons can freely travel around by moving in and out of the empty sites. In this organic material, however, strong repulsion between the electrons in the full

and empty states suppresses free movement. The layered structure and arrangement of molecules into layered, triangular patterns (see image) removes the freedom of the electrons' spin such that the molecules line up and form valence bonds.

The only way for electrons to break free is to forcefully add additional electrical charge to the system, or to subject the material to high pressure. In both cases, the electronic states change such that the material undergoes a Mott transition to a conducting state.

Studying the Mott transition with the precision required to unravel fundamental physical properties, however, is particularly challenging. Therefore, rather than relying solely on electrical conductivity measurements, the researchers combined these observations with thermoelectric power measurements, which track changes in electrical behavior with temperature. The experimental data clarified previous conflicting experimental results and revealed that the Mott transition belongs to a universal class of phase transitions.

Although this discovery provides a deeper understanding of Mott transition materials, many specific properties of $EtMe_3P[Pd(dmit)_2]_2$ remain to be determined, notes Abdel-Jawad. "For example, we do not fully understand the conductivity of the insulating side of the Mott transition." Additional experiments on this unusual compound therefore promise the discovery of further intriguing physics beyond the phase transition itself. ■

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MATERIALS | PRESS RELEASE

Upside-down solar cells exhibit high performance

Flexible polymer solar cells have been fabricated with an efficiency close to that needed for commercial viability

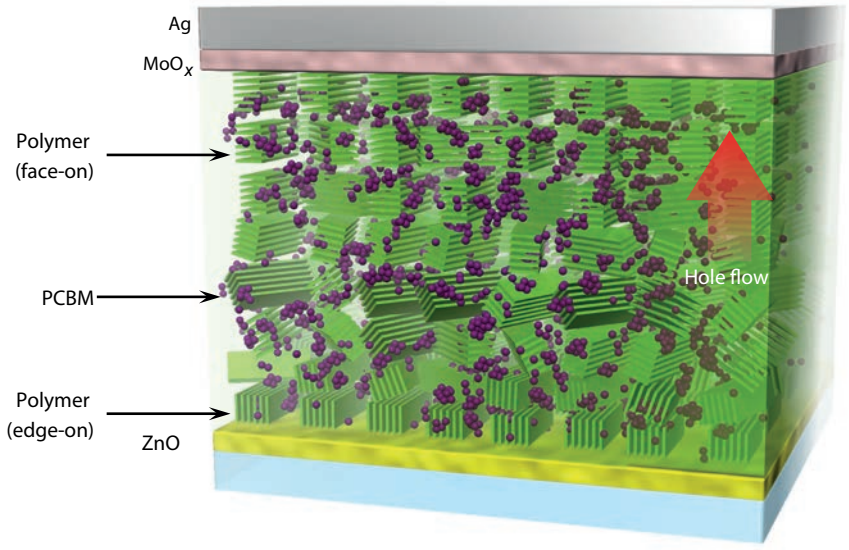
Using carefully designed materials and an 'inverted' cell architecture, RIKEN scientists have fabricated polymer solar cells with a power conversion efficiency—a measure of how effective solar cells are in converting sunlight into electricity—of 10 per cent, bringing these cells close to commercial viability[1].

Polymer solar cells could become a viable alternative to conventional solar cells that have silicon substrates. They are made of inexpensive and flexible polymers, and they can be deposited on glass or plastic substrates, allowing the construction of large-scale structures. While they are cheaper to manufacture and more environmentally friendly than their silicon counterparts, they currently have lower power efficiencies.

Itaru Osaka and Kazuo Takimiya of the RIKEN Center for Emergent Matter Science and their co-workers have now fabricated a type of polymer solar cell called a bulk-heterojunction solar cell—where the electron-donor and electron-acceptor layers are mixed together (see image). Their solar cell had a high power conversion efficiency of 10 per cent—close to that required to make these materials commercially viable.

"We began experimenting with a substance we had previously developed," explains Osaka, "and were able initially to achieve a power conversion efficiency of about 8 per cent, with a fairly thick active layer."

Surprisingly, the researchers found that the conversion efficiency went up to about 10 per cent when they used an inverted architecture, in which the light enters through a transparent negative electrode.



Schematic illustration of an inverted solar cell.

"This is abnormal for cells of the type we built," says Osaka. "We believe that it is due to the alignment of molecules inside the mixed layers."

The scientists analyzed the composition of the materials using the SPring-8 synchrotron facility and found that in the inverted model, the orientation of the molecules in the active layer was very commonly 'face-on', an orientation well suited to the transport of electron 'holes' through the material. "We surmised that this was the secret to the success in the experiment." Takamiya says. "It turns out that by trying something that might seem unusual, we got a surprising result, and through this we were able to understand something about what makes cells more or less efficient."

"This is an exciting result because we now have an understanding of how we can move forward to create polymer solar cells with greater efficiency," Takamiya adds. "We hope that researchers around the world will be able to make use of these results to create commercially viable cells." ■

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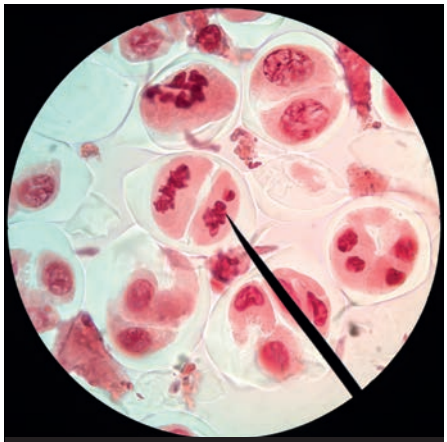
BIOLOGY

Cells' universal law of change

Analysis of gene expression in growing cells reveals a universal law that governs intracellular changes

The internal workings of a cell involve many molecular components, and the ability to alter all of these components in tandem enables cells to adapt to different conditions. Research by Chikara Furusawa from the RIKEN Quantitative Biology Center and colleagues has now advanced our understanding of how these global changes are controlled by identifying a universal law that governs changes in gene expression as cells grow[1].

Furusawa, in collaboration with Kunihiko Kaneko from the University of Tokyo and Tetsuya Yomo from Osaka University, used a technique called transcriptome analysis to measure the expression of thousands of genes across the genome in an attempt to reveal global changes in gene expression. They restricted their analysis to cells with a steady growth rate. This simplified model was based on the assumption that in order to maintain a steady rate of growth, cells must maintain their internal state by globally increasing gene expression.



A universal law governs the changes in gene expression as cells divide and grow.

This transcriptome analysis enabled the researchers to derive a formula that mathematically describes the global change in gene expression as cells grow. Specifically, they found that the change in the concentration of any intracellular component is related to the overall growth rate of the cell: a cell undergoing steady growth must sustain each of its internal components such that the abundance of all components doubles during each cell-division event.

"During adaptation and evolution, the changes in expression of all genes are highly correlated with one another, determined by a common proportion coefficient that is determined by the growth rate of the cell," explains Furusawa. "Even though there is a huge number of molecular species inside cells, their change is constrained by this very simple law."

The team tested their theory against real data for the commonly studied bacteria *Escherichia coli* and found good agreement, demonstrating that their model is biologically sound. Although restricting the model to cells that are growing steadily limits its applicability, Furusawa believes the technique holds promise.

"There are states in which cells do not grow, and ways in which we can characterize such dormant states remain to be discovered," he explains. "We need to expand our theory to complex biological systems."

The researchers are now investigating the evolution of *E. coli* under various environmental conditions, and they intend to apply the theory to describe the dynamics of adaptive evolution with several macroscopic variables.

"In the future, this approach might enable us to predict changes in adaptation and evolution, which is useful for engineering and medicine," says Furusawa. ■

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Synchrotron radiation

Brighter, shorter x-ray pulses to share

From a small group of pioneering researchers using the radiation wasted during high-energy physics experiments has grown an entire scientific field dedicated to constructing synchrotron radiation facilities. The demand for the most brilliant source of x-rays from researchers of every hue has been relentless. The RIKEN SPring-8 Center is addressing this need by making some of the most advanced x-ray sources accessible to them all.

Tetsuya Ishikawa is director of the RIKEN SPring-8 Center
Ishikawa joined RIKEN in 1993, became chief scientist at the Coherent X-Ray Optics Laboratory in 1998 and director of the RIKEN SPring-8 Center in 2006. In 2012, Ishikawa received the Medal of Honor with Purple Ribbon from the Government of Japan for his enormous contribution to x-ray optics.

Synchrotron radiation was quite a nuisance to high-energy physicists. They spent the last half of the last century investigating the basic building blocks of matter by first accelerating particles—the faster, the better—and then smashing them into other particles. Large donut-shaped facilities called synchrotrons made it possible to accelerate charged particles like electrons almost to the speed of light by making them undergo continuous revolutions around a ring of magnets.

But when charged particles are forced to speed up, slow down or change direction, they emit electromagnetic radiation or light. At slower speeds, this radiation appears in the form of weak, long, low-frequency waves moving in almost every direction. As a particle is accelerated to nearly the speed of light, it produces a spectacular beam of light whose wavelengths span a large portion of the spectrum, extending all the way up to the ultraviolet and x-ray regions.

For decades, this so-called 'synchrotron' radiation represented the single largest source of energy loss in high-energy physics experiments. Then a group of researchers in the 1960s started making use of the waste product parasitically, which led to an explosion in x-ray science.

Whipping x-rays

The resolution of observations using light is limited by the wavelength. In the visible regime, for example, wavelengths are just under a micrometer long, and can therefore be used in microscopes to observe the micro-world. X-rays, with wavelengths more than a thousand times shorter, allow us to see the nano-world, from individual atoms to twisting DNA and carbon nanotubes. Prior to synchrotron radiation, however, the only x-ray sources were x-ray tubes, which involved slamming high-speed electrons against a metal target. First demonstrated by Wilhelm Röntgen in 1895, the x-rays produced using this method could not be tuned to a desired wavelength and fell far short of the brilliance of synchrotron radiation, which was developed subsequently.

By the 1980s, researchers, especially those in the fields of spectroscopy and crystallography, had lobbied for facilities solely designed to generate synchrotron radiation. In these second-generation synchrotrons, bunches of electrons confined in a central storage ring emit x-rays into beamlines. At the end of each beamline is a tiny sample waiting to be hit, and a scrutinizing researcher.

Synchrotrons saw major upgrades in the 1990s, with the construction of three mega-facilities: the European Synchrotron Radiation Facility in France, the Advanced Photon Source in the United States and, finally in 1997, SPring-8 in Japan. These third-generation synchrotrons were fitted with straight sections in which undulators were installed. The undulators consisted of two parallel rows of magnets having alternating polarities through which electrons undulate like tiny ripples to produce brilliant x-rays.

The brightest light of them all was generated by SPring-8. Established through collaboration between RIKEN and the former Japan Atomic Energy Research Institute, the storage ring at SPring-8 has the largest circumference (1,436 meters) and can maintain the highest electron energy (8 gigaelectronvolts) of all three facilities. This means that it can penetrate deep into heavy atoms like uranium and plutonium. But the crucial innovation is in its insertion devices, where the parallel rows of magnets are brought inside an ultrahigh vacuum chamber, closer to the zigzagging electron beam to produce even more brilliant radiation.

Soon after the completion of SPring-8 came another major breakthrough with the construction of the SPring-8 Angstrom Compact free electron LAser (SACLA) in 2011. The in-vacuum undulator technology was again adopted in SACLA, which has a linear structure (Fig. 1). The radiation is emitted as a single coherent beam of x-rays—a distinguishing characteristic of lasers—in short pulses so powerful that they destroy the sample. SACLA's x-ray laser is a billion times more brilliant and ten thousand times shorter than the radiation emitted at SPring-8.

SACLA is one of only two free-electron lasers in the world capable of producing hard x-rays with wavelengths about a tenth of a billionth of a meter (a tenth of a nanometer) long—the other is the Linac Coherent Light Source (LCLS) at the SLAC National Accelerator Laboratory in the United States. Thanks to RIKEN's in-vacuum undulator technology, SACLA requires much less energy than the LCLS (8 versus 14 gigaelectronvolts) to produce a shorter wavelength x-ray laser (0.06 versus 0.1 nanometer). And the total length of the SACLA facility at 700 meters is considerably shorter than the two-kilometer facility in the United States. Indeed, SACLA is the first compact x-ray laser source[1].

Figure 1: Thanks to RIKEN's in-vacuum undulator technology, the SPring-8 Angstrom Compact free electron LAser (SACLA) at RIKEN can emit a coherent x-ray laser beam with wavelengths about a tenth of a nanometer long.

Protein pumps and photosynthesis

With these advances in the production of synchrotron radiation a realization has emerged among scientists of its potential for interdisciplinary applications. Large facilities like SPring-8 and SACLA have made it possible for researchers of various scientific backgrounds to access the light

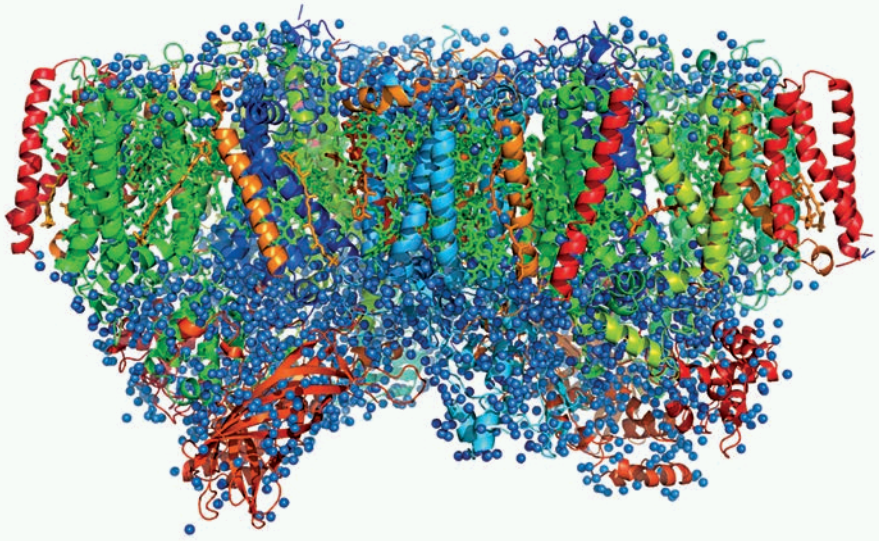

Figure 2: The three-dimensional structure of photosystem II was determined using an ultrashort laser pulse with a wavelength of less than 0.2 nanometers produced by SACLA.

source. Close to 60 beamlines tap into SPring-8's storage ring, and nearly half are publicly accessible to researchers from any institution. The facility welcomes about 20,000 researchers each year, adding up to more than 180,000 users over the past two decades of operation.

The results have been tremendous, spawning more than 10,000 papers in a wide range of fields, including life sciences, materials science, Earth science and nuclear physics.

One group of researchers from the University of Tokyo, for example, has crystallized the calcium pump proteins embedded in muscle cell membranes that induce muscle motion. By analyzing the patterns of x-ray diffraction from their samples at SPring-8, the researchers were able to determine the three-dimensional structures of these protein pumps, at various stages of calcium attachment and non-attachment.

Tokyo Institute of Technology researchers have also collaborated with SPring-8 to recreate the structure of a mineral found 2,900 kilometers underground where pressures reach above 125 gigapascals and temperatures above 2,000 degrees Celsius.

Coming full circle to high-energy physics, an international team of physicists discovered a subatomic particle consisting of five of the most fundamental elementary particles called quarks (in contrast, protons and neutrons are made of only three quarks).

Some of the research at SPring-8 has had direct industrial benefit, such as the measurement of silica nanoparticles found in rubber, which led to the commercialization of low-resistance automobile tires that improve fuel efficiency by approximately 6 per cent.

SACLA has enabled researchers to further refine their scope of investigation, from molecules arranged periodically in a crystal to whole, stand-alone molecules. And by producing intense laser pulses every one-sixtieth of a second, researchers can capture events as fleeting as changes in the bonds holding atoms together to create molecular movies.

Researchers at RIKEN and Okayama University have used SACLA to determine the first high-resolution crystal structure of an undamaged protein called photosystem II (Fig. 2)[2] that generates the oxygen we breathe via the photosynthetic process. And SACLA successfully induced a nonlinear optical effect in which two lower-frequency x-ray photons are absorbed in a crystal of pure germanium and emitted as one higher-frequency photon. More than 2,000 researchers have used SACLA since its two beamlines opened to the public in March 2012. By popular demand, RIKEN started operation of another beamline in April 2015.

Brighter synchrotron radiation

Though sometimes located within walking distance from each other, free-electron lasers and synchrotron facilities currently exist in leagues of their own. The next frontier in x-ray science will be to produce a light as coherent as SACLA but that has mild to moderate intensity so as not to damage the target. This can be achieved within a decade by upgrading the original synchrotron design. RIKEN is taking the first steps in this direction with a scheduled upgrade of SPring-8 to be completed by the early 2020s.

SPring-8-II will rely on a revolutionary technology called the multi-bend achromat that can further concentrate the electrons in the storage ring to produce more coherent light. Current storage rings use double-bend achromats made of two magnets per unit. SPring-8-II will have five-bend achromats containing five magnets per unit to emit an x-ray beam that is 60 times more brilliant.

Brighter and more coherent sources of x-ray radiation will continue to be sought, but the real challenge remains satisfying the voracious appetite for the technology. By delivering faster results, SPring-8-II will be able to shorten user turnaround. And RIKEN plans to construct SACLA-2 that will be ten times shorter than SACLA, and could be reproduced in universities and research centers across Japan. The construction of a mid-sized synchrotron facility is also under discussion. These developments will allow researchers as far afield as the social sciences to reap the rewards of synchrotron radiation. ■

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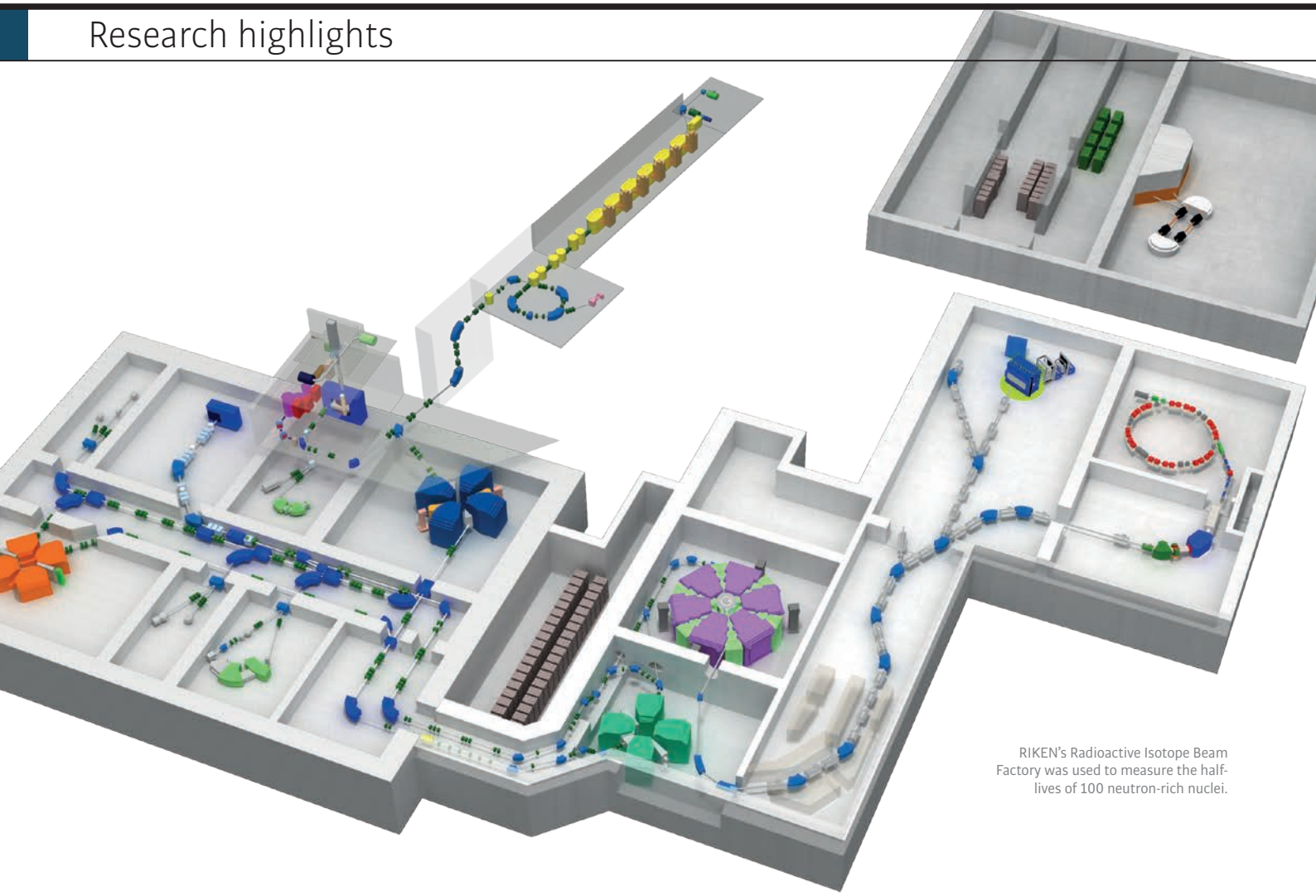

RIKEN's Radioactive Isotope Beam Factory was used to measure the half-lives of 100 neutron-rich nuclei.

PHYSICS | PRESS RELEASE

Nuclear half-lives affect birth of new elements

Measurements of the half-lives of short-lived nuclei shed light on the elemental composition of the Universe

Using RIKEN's Radioactive Isotope Beam Factory (see image)—one of the world's most powerful devices for creating exotic atomic nuclei—RIKEN scientists have, with international collaborators, precisely measured the half-lives of 110 nuclei, 40 of which had never been measured before[1]. These nuclei are located at the boundary of the known nuclear chart and despite having short lifetimes—measured in milliseconds—they imprint their properties on the chemical composition of the Universe.

The team generated exotic nuclei by colliding a uranium beam with a beryllium target. They then measured the half-lives of the produced nuclei by measuring beta decay and gamma emission.

This study is a major step toward providing an experimental ground for models of the mysterious astrophysical 'r-process', which is believed to be responsible for creating many of the elements in the Universe heavier than iron.

The r-process occurs only under extreme conditions, such as when a supernova core collapses to become a neutron star, releasing tremendous energy in the process. Under those extreme neutron-rich conditions, atomic nuclei absorb neutrons to become increasingly heavy, and then undergo beta decay, leaving the nucleus one element higher in the periodic table. In this way, nuclei gradually creep up the periodic table and create new elements.

Hundreds of isotopes participate in this process, which occurs extremely rapidly. Without accurately knowing their

half-lives—how quickly they cast off an electron—it is difficult to effectively model the violent processes that created the elements we see today. While significant progress has been made, current models cannot fully explain the elemental abundances of stars.

"It was very exciting to explore unknown territory in the nuclear chart and discover the half-lives of isotopes that had never been measured before," says Giuseppe Lorusso of the RIKEN Radioactive Isotope Physics Laboratory. "This new data allows us to get closer to understanding the mystery of nucleosynthesis."

While the results indicate that current models may be capturing the essential physics of the r-process, they also brought a surprise.

"We found that after reducing the uncertainties of nuclear physics with our measurements, the difference in the abundances of elements such as tin, antimony, iodine, and cesium, among very old stars created in the early Universe can be understood as originating from differences in the r-process conditions," Lorusso explains.

"This opens up the possibility that by looking at the distribution of elements in those stars, we can gain an understanding of the precise environment in which the r-process took place." ■

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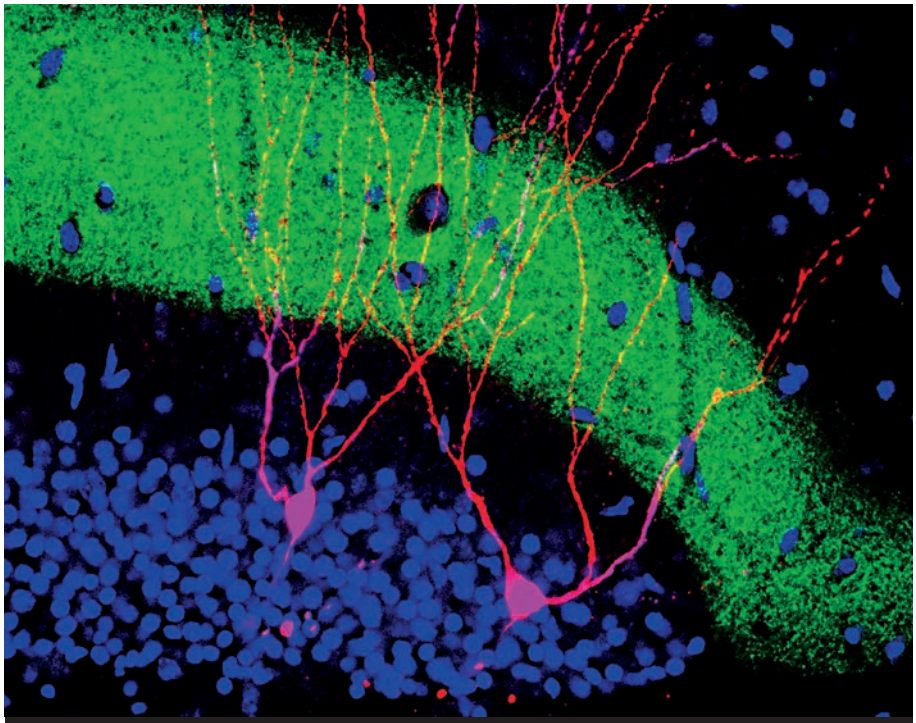
BIOLOGY | PRESS RELEASE

Retrieving 'lost' memories

Mice suffering from amnesia can recall past memories when specific neurons are artificially activated

Retrograde amnesia, the inability to recall established memories, is associated with traumatic brain injury, Alzheimer's disease and other neurological conditions. Whether memories lost to amnesia are completely erased or merely irretrievable, however, remains an open question. Now, researchers from the RIKEN–MIT Center for Neural Circuit Genetics have demonstrated that traces of old memories remain in the brains of amnestic mice, and that the cellular pathways underlying them can be reactivated, allowing lost memories to retrieved[1].



Two cells (stained pink) in the mouse hippocampus; the one on the right was involved in a memory engram.

The research team, led by Susumu Tonegawa, director of the RIKEN Brain Science Institute, was interested in how stable memories are formed in the brain and whether memories whose storage was disrupted by chemically induced retrograde amnesia could be recalled. "Brain researchers have been divided for decades on whether amnesia is caused by an impairment in the storage of a memory, or in its recall," says Tonegawa.

The researchers trained mice to associate a mild foot shock with a specific environment, chamber A, eliciting a typical 'freezing' behavior. Eventually, trained mice would freeze in chamber A even without the shock. Neurons activated during memory formation were genetically labeled to allow them to be visualized and reactivated. Some of the mice were then given a chemical that induces retrograde amnesia, while the other mice received saline as a control.

As expected, when the amnestic mice were returned to chamber A they did not freeze, indicating that they could not recall the association between the chamber and the mild foot shock. The researchers then used optogenetic technology to selectively activate neurons that were genetically labeled during the training in

chamber A with a blue-light-sensitive protein, but this time while the mice were in a new, neutral environment, chamber B.

Surprisingly, during activation of the cells involved in the foot shock memory, collectively called a 'memory engram', with blue light pulses, the amnestic mice froze just as much as the control mice. This indicates that they remembered they had acquired the memory, even though they could not recall it when placed in chamber A. To explain these findings, the researchers suggest that different processes may control memory encoding and recall.

"Our conclusion is that in retrograde amnesia, past memories may not be erased, but could simply be lost and inaccessible for recall," says Tonegawa. "These findings provide striking insight into the fleeting nature of memories, and will stimulate future research on the biology of memory and its clinical restoration." ■

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BIOLOGY

A survival-promoting protein with a double edge

A signaling protein that sustains immune defenses could also promote cancer

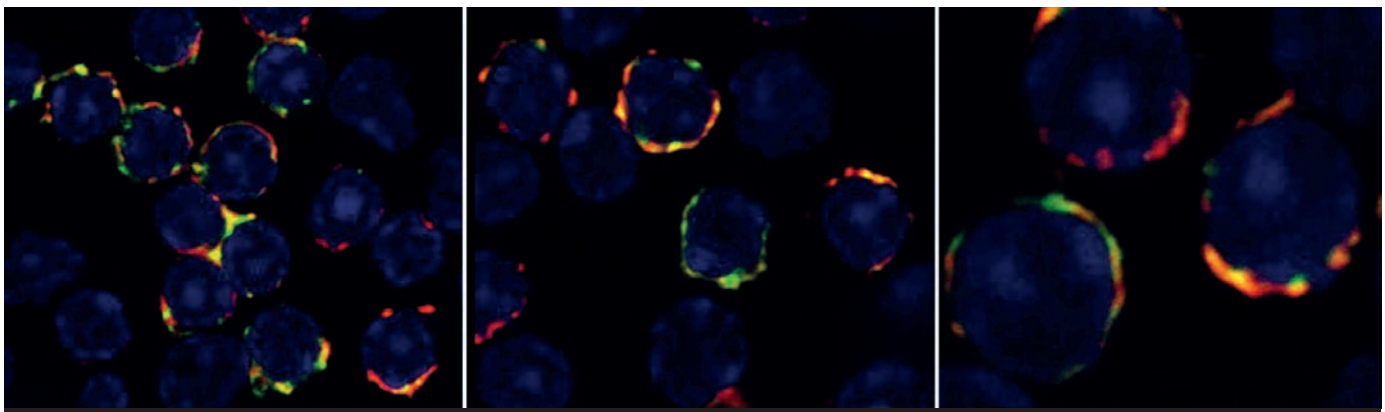
A recently discovered immune system protein called FcμR could be involved in various autoimmune diseases and may actually contribute to certain cancers, a collaborative study by researchers from RIKEN, Fudan University in China and the Tokyo Medical and Dental University has found[1].

The immune system maintains reservoirs of antibody-producing peripheral B cells within the spleen and lymph nodes, which are kept at the ready in case a targeted immune response is required. Specific 'survival signals' are used to sustain these cell populations both at rest and after B cells have been activated by an antigen. "FcμR is a recently discovered B-cell protein that we found plays a critical role in immune response by supporting B-cell survival," says Rika Ouchida from the RIKEN Research Center for Allergy and Immunology. "But exactly how it does this has remained unclear."

B-cell activation occurs through a protein complex called the B cell receptor (BCR; see image). Ouchida and her co-workers confirmed that FcμR boosts survival in the aftermath of B-cell stimulation, and that it does so via direct interaction with the BCR complex.

Previous investigations of the immune system had shown that peripheral B-cell survival is coordinated through the joint activity of BCR and another protein called BAFFR. These two proteins work together to trigger parallel signaling pathways that switch on key survival genes. Ouchida and her co-workers found a similar relationship between BCR and FcμR, with one notable difference—BAFFR can trigger some signaling activity by itself, whereas FcμR cannot. The team suggests that this may be related to BAFFR's role in keeping inactive peripheral B cells alive, whereas FcμR acts primarily in antigen-stimulated peripheral B cells. "This means that FcμR contributes to the support of long-term B-cell survival during the immune response," says Ouchida.

BCR does not just generate survival signals; it can also help eliminate B cells that recognize the body's own proteins. The researchers



Fluorescence microscopy images showing the co-localization of FcμR (red) and immunoglobulin M (green), which together form the B cell receptor (BCR) complex.

observed more of these self-targeting antibodies in FcμR-deficient mice, suggesting a possible role for this protein in preventing autoimmune diseases.

This same survival pathway, however, could also be exploited by certain blood cancers. "We know that FcμR levels are much higher in malignant B cells from patients with chronic lymphocytic leukemia," says Ouchida. "Our results suggest that elevated FcμR expression may enhance survival and contribute to the pathogenesis of these malignant cells."

In future work, Ouchida and her colleagues will attempt to dissect the molecular mechanisms of FcμR associated with various disease states. ■

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BIOLOGY | PRESS RELEASE

Why human egg cells don't age well

Premature separation is found to be the primary cause for chromosomal errors in older eggs

When egg cells form with an incorrect number of chromosomes—a problem that increases with age—the result is usually a miscarriage or a genetic disease such as Down syndrome. Now, researchers at the RIKEN Center for Developmental Biology have pinpointed a significant event that leads to these types of age-related chromosomal errors[1]. They discovered that as egg cells mature in older women, paired copies of matching chromosomes often separate from each other prematurely, leading to early division of chromosomes and their incorrect segregation into mature egg cells.

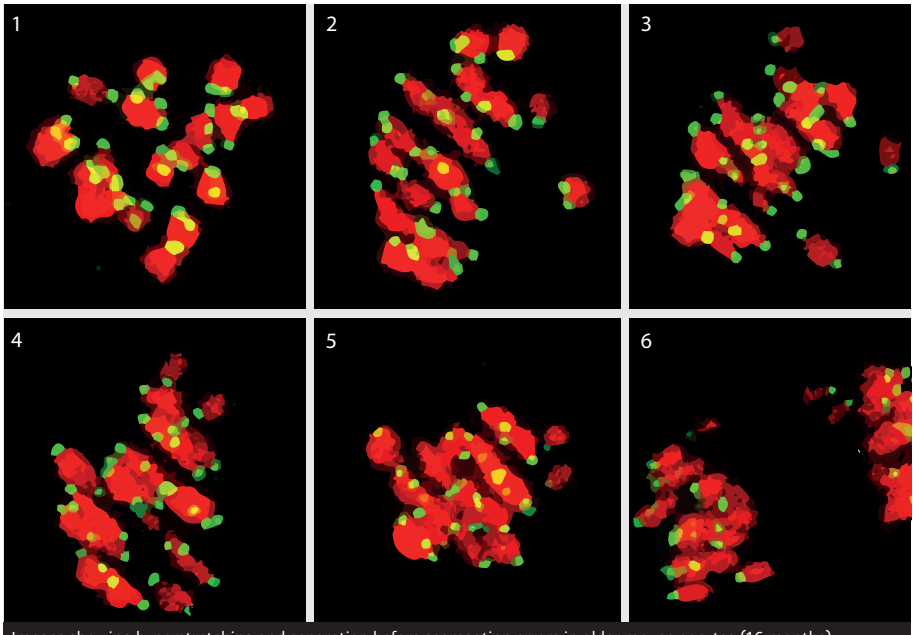
Most cells have two copies of each chromosome—one from each parent. Immature egg cells begin this way, but are transformed through a process called meiosis into mature egg cells that have only one copy of each chromosome. At the beginning of meiosis, each chromosome copies itself and joins with its matching pair to form a group of four chromosomes that swap genetic material.

These groups of four chromosomes—called bivalents—split into single pairs, and the cell divides. One part continues as the egg cell and the other degrades. In the second stage of meiosis, the single pairs of chromosomes separate and the egg cell divides again in the same way, leaving a single mature egg cell with one copy of each chromosome.

"We found that in older cells, the bivalents sometimes separate early, and this leads to division of sister chromatids in the first stage of meiosis, rather than in the second stage," explains team leader Tomoya Kitajima.

To determine the most common type of age-related segregation errors, the researchers used high-resolution imaging to visualize chromosomes in live mouse egg cells during the first stage of meiosis. They found that chromosomes were always correctly distributed in young egg cells, whereas a little under 10 per cent of older cells suffered from segregation errors. The chromosome-tracking data revealed that the dominant error was predivision of sister chromatids.

The researchers confirmed that the number of singly paired chromosomes was higher in



Images showing hyperstretching and separation before segregation errors in older mouse oocytes (16 months). Chromosomes are seen as red, and kinetochores are green.

older mouse and human egg cells, indicating that age-related segregation errors could be tracked to increased numbers of prematurely separated chromosome pairs.

"We were surprised and pleased that the vast majority of errors are preceded by a single common event—bivalent separation," says Kitajima. "Now we can focus our efforts on developing an artificial tie to suppress premature separation and on understanding the molecular mechanism underlying the age-related reduction in bivalent cohesion that appears to precede it." ■

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PHYSICS

Electrons reveal the hidden structure of proteins

An innovative crystallography technique uncovers the charge distribution in protein structures

An electron crystallography technique developed by RIKEN and the University of Tokyo makes it possible to probe hidden details of small three-dimensional protein crystals as a potent new tool for studying the function and structures of biological macromolecules[1].

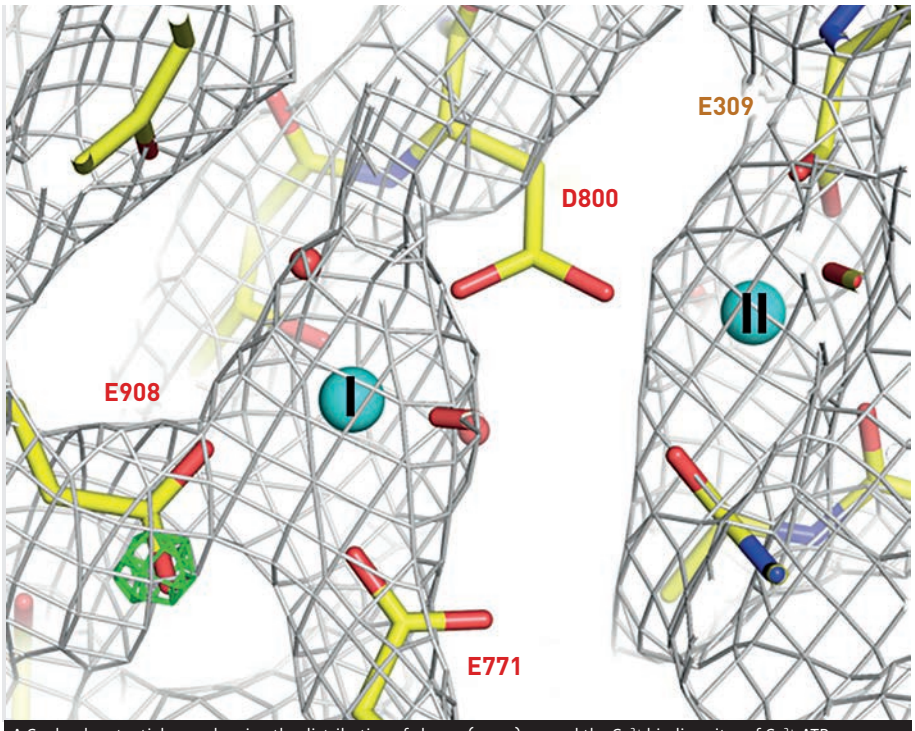
The atomic structure of crystalline materials is routinely determined by a technique called x-ray crystallography, in which the scatter of x-rays is used to identify the location of atoms in the crystal. This method can be used to reveal the structures of many proteins, but for macromolecular complexes and proteins that occur in cell membranes, it is often very difficult to obtain crystals large enough and yielding clear enough diffraction patterns to be able to determine their structure using even the most intense x-rays.

Electron crystallography is based on a similar principle but uses a beam of electrons instead of x-rays and has the advantage of being 100,000 times more strongly scattered by organic molecules. This makes it possible to use much smaller and thinner crystals, which are all that are available for many biologically important proteins. Electron crystallography can also resolve charge states, revealing critical information about protein function. However, the electron beam can damage the sample, and collecting and analyzing data from crystals a few molecular layers thick is challenging.

Koji Yonekura from the RIKEN SPring-8 Center and Chikashi Toyoshima from the University of Tokyo led a collaborative study to develop an electron crystallography method that overcomes these limitations through the use of a new diffractometer and novel data collection and processing techniques. "As no technology was available for electron crystallography of such thin crystals," says Yonekura, "we had to develop both the hardware and software, in many cases from scratch."

To reduce radiation damage, the new diffractometer operates at cryogenic temperatures. To obtain useful structural information from the weak electron scattering signals, the diffraction spots produced by scattered electrons are recorded and stacked while the crystal sample is rotated. A new data analysis package then produces a precise atomic model that includes charge information.

The team tested the technique on ultrathin crystals of two common enzymes, Ca^{2+}-ATPase and catalase, producing 'Coulomb potential' maps showing the charge states of ion and



A Coulomb potential map showing the distribution of charge (cages) around the Ca^{2+} binding sites of Ca^{2+}-ATPase.

amino acid residues at the enzymes' active sites (see image).

These maps, note Yonekura, show key aspects of the working mechanism of the biological macromolecules. "Using this technique we have resolved the charge states of ions and amino acids—this information cannot be obtained using other techniques." ■

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BIOLOGY

Multicolored nano-lanterns light up cells

Proteins that glow yellow, orange or cyan can be used to illuminate a range of biochemical processes without light excitation

Fluorescent proteins are invaluable tools for studying biological processes, but they only glow when stimulated with an external light source, which can damage cells or trigger unwanted biochemical reactions. A RIKEN-led research team has now developed an alternative imaging technique using luminescent proteins called 'nano-lanterns' that are powered by chemical energy rather than light[1].

Previous attempts to use luminescent proteins for imaging foundered because their light is usually too dim to track rapid changes inside cells. Instead, Yasushi Okada, Akira Takai and colleagues from the RIKEN Quantitative Biology Center, in collaboration with the team of Takeharu Nagai from Osaka University, looked to nature for inspiration.

The sea pansy *Renilla reniformis* contains an enzyme called *Renilla* luciferase (RLuc) that helps to oxidize a molecule called coelenterazine—a chemical reaction that produces a flash of blue light. A few years ago, Nagai's team coupled a mutant form of RLuc with a yellow-green fluorescent protein called mVenus to produce a luminescent imaging agent.

Building on this work, Okada and his colleagues created two more coelenterazine-oxidizing luminescent agents using the same approach by coupling RLuc with cyan and orange fluorescent proteins. The resulting nano-lanterns are about 20 times brighter than natural RLuc. "The luminescent probes are bright enough to be detected with an iPhone camera," says Okada.

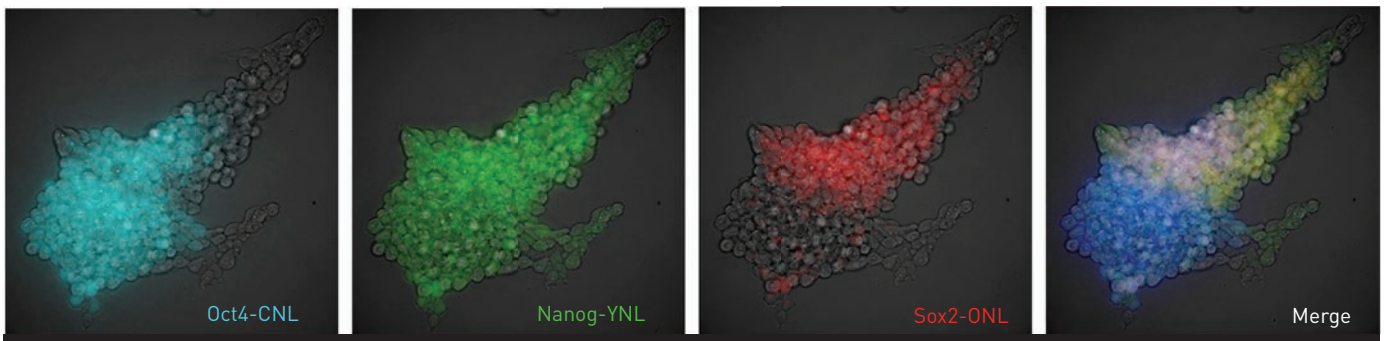
To demonstrate their technique, the team attached nano-lanterns to cellular structures such as lysosomes to track their movements over several minutes. They also monitored the concentration of calcium ions in cells, using nano-lanterns with a mutant RLuc that was split in two such that the lanterns only shone when a calcium ion joined the two halves together.

Finally, the researchers used their nano-lanterns to study gene expression. By adding the genetic codes for the nano-lanterns next to three different genes inside mouse cells, the team was able to produce a distinct color signature when those genes were expressed. In just ten seconds, this revealed patterns of gene expression across a colony of cells (see image).

The nano-lanterns are still about 100 times dimmer than conventional fluorescent probes, and rely on a steady supply of coelenterazine to keep glowing, but Okada believes that these limitations are challenges that will be overcome. "Another goal is to create red nano-lanterns that could be useful for deep-tissue imaging," he notes. The team is now developing microscopes for super-resolution luminescence imaging, and using the nano-lanterns to further analyze gene expression within single cells. ■

Reference

1. Takai, A., Nakano, M., Saito, K., Haruno, R., Watanabe, T. M., Ohyanagi, T., Jin, T., Okada, Y. & Nagai, T. Expanded palette of Nano-lanterns for real-time multicolor luminescence imaging. *Proceedings of the National Academy of Sciences USA* **112,** 4352–4356 (2015).



Each of the nano-lanterns highlights the expression of different genes in mouse embryonic stem cells.

RIKEN SPring-8 Center

Energizing photons to the max

The RIKEN SPring-8 Center hosts the most powerful synchrotron radiation facility in the world and an x-ray free-electron laser that is a billion times brighter

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Encircling mount Miharakuriyama, about 100 kilometers west of Osaka, is a 1,436-meter-long obstacle course for subatomic athletes. Here, in the SPring-8 synchrotron facility at the RIKEN SPring-8 Center, bunches of electrons wiggle, lurch and careen through vacuums and magnetic fields at 200,000 loops a second. They whiz past, like Sonic the Hedgehog, in a flattened mass of ten billion electrons, three hairbreadths wide and nearly half a centimeter long. Just next door is the SPring-8 Angstrom Compact free electron LAser (SACLA), where particles ricochet like speeding bullets through a hundred-meter barrel of magnets in femtosecond rounds.

These electrons are accelerated to nearly the speed of light to produce some of the brightest electromagnetic radiation or light in the world, especially in the x-ray region. Researchers of all types use them to image objects even smaller in size than an atom.

The SPring-8 Angstrom Compact free electron LAser (SACLA) is only walking distance away from the SPring-8 Center's synchrotron radiation storage ring (above). SACLA experiments are performed in the shared experimental hall (opposite page).

Custom beamlines

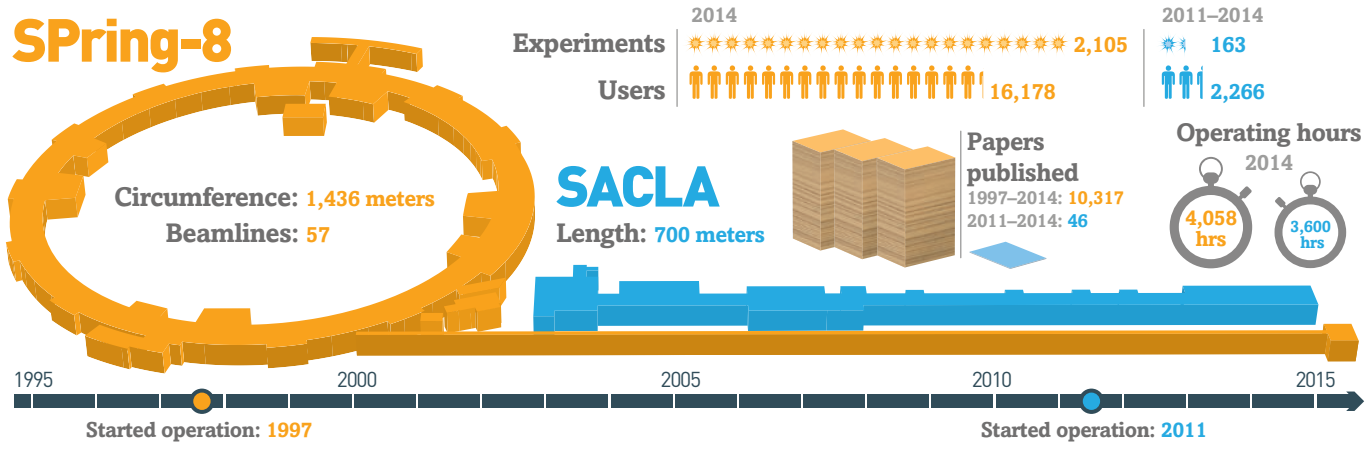
Precision is critical at the RIKEN SPring-8 Center. The synchrotron storage ring consists of a chain of 1,500 massive magnets. To prevent the accelerator from expanding and contracting with changes in temperature, it is housed in a frame decoupled from its outer building. And the building itself has been carved into the bedrock to prevent the gravitational pulls of the Sun and Moon, which deform the Earth's surface, from altering the length of the racetrack. Monitors installed at 300 locations along the rim detect any micrometer-long deviations that may develop over the course of a day.

Scientists and engineers at SPring-8 have constructed 57 beamlines that extract synchrotron radiation from SPring-8, and three beamlines at SACLA that produce coherent x-ray laser beams. The beams have been customized to suit the needs of research fields including biology, chemistry, photonics, quantum dynamics, magnetic materials and the physics of systems under extreme temperatures and pressures. Almost half of the beamlines at SPring-8 are publicly accessible to researchers from any institution and a fifth are used by industry.

Lighting the way

These facilities have yielded illuminating results, with about 800 papers based on experiments conducted at SPring-8 published annually. In 2015, for example, RIKEN researchers observed a polymer-based solar cell under synchrotron radiation to discover the secret of its high power conversion efficiency. And in 2014, among other discoveries at SACLA, researchers revealed the internal structure of living *Microbacterium lacticum* cells sealed in a liquid solution between two synthetic membranes. The laser beam was directed at a micrometer-sized spot, and snapshots of the cells were taken just femtoseconds before they died of radiation damage.

Highlighting the benefits of synchrotron radiation is another important aspect of the center's work. For example, for the past eight years, RIKEN has been hosting the Cheiron School, which informs graduate students, postdoctoral researchers and young engineers in the Asia-Pacific region about useful techniques and applications. Over 60 participants are expected to participate in the next session in September 2015 and afterwards spread the knowledge of a powerful means to see the world.



Interns studying at the RIKEN Yokohama Campus, together with Piero Carninci (top row, left), deputy director of the RIKEN Center for Life Science Technologies (CLST), and Aki Minoda (top row, right), unit leader at the CLST.

Programs for Junior Scientists

RIKEN strives to provide the best and most exciting opportunities for young scientists from all over the world in the crucially important early stages of their careers. The Special Postdoctoral Researcher Program offers creative young scientists the chance to participate in autonomous and independent research under the direction of a RIKEN laboratory head. The Junior Research Associate program provides part-time positions for enthusiastic and open-minded young researchers enrolled in doctorate programs at Japanese graduate schools. RIKEN also accepts non-Japanese PhD candidates as International Program Associates through a cooperative undertaking of students between RIKEN and collaborating universities.

Internship and summer schools

"Should I get a job or should I go to graduate school and get a PhD?" This is one of the biggest life-changing questions that young scientists face. We strongly believe that even undergraduate students should be given a chance to experience a research environment.

The RIKEN Brain Science Institute gives young researchers a stimulating opportunity to study brain science. At the Nishina School, students from selected universities can learn about theoretical and experimental nuclear physics at the RIKEN Nishina Center for Accelerator-Based Science. The RIKEN Center for Integrative Medical Sciences offers students the chance to learn about recent research in immunology. And with the help of the Asia–Oceania Forum for Synchrotron Radiation Research, RIKEN conducts the Cheiron School, where participants can learn about synchrotron radiation science at SPring-8, the world's largest third-generation synchrotron facility.

Also, the RIKEN Center for Life Science Technologies hosted many undergraduate interns from various countries this summer. These interns contacted us directly or through their supervisors to ask for internship opportunities. From our experience launching the international FANTOM (Functional Annotation of the Mammalian Genome) consortium, we have learned that promoting friendship among scientists all over the world, including upcoming young researchers, is critical for conducting mutually fruitful research.

Even if you do not find a suitable program at RIKEN, don't give up—we are always open to new ideas. Please contact us and ask us for a chance to become an intern at RIKEN.

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Harima

SPring-8 Center
SPring-8 Synchrotron Radiation Facility
SACLA X-ray Free Electron Laser Facility

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