World Premier International Research Center Initiative (WPI)
Executive Summary（Interim Evaluation）

Host Institution	University of Tsukuba	Host Institution Head	Kyosuke Nagata
Research Center	International Institute for Integrative Sleep Medicine	Center Director	Masashi Yanagisawa

*Summarize the Self-Evaluation Report for Interim Evaluation (**within 4 pages** including this page).

1. Summary of State of WPI Center Project Progress

Research activities

Sleep is an essential behavior that everyone experiences daily and it takes up one third of one's entire lifetime, although the very fundamental mechanisms of sleep and its *raison d'être* remain still unknown. While sleep has been a complete black box, its medical and social importance is very clear. Healthy sleep is necessary for maintaining our mind and body fitness; lack of sound sleep not only causes a reduction in higher brain functions including memory and decision making, but increases the risk of mood disorders such as depression as well as metabolic syndrome. Domestic economic loss caused by sleep disorders is estimated as 3.5 trillion yen/year, hence it is the urgent need to solve sleep-related issues.

We set out our major research objectives to solve the issues of sleep disorders as follows.

1) To elucidate the fundamental mechanisms of sleep/wake regulation
2) To elucidate molecular pathogenesis of sleep disorders and related diseases
3) To develop treatments for sleep disorders

To achieve these objectives, wide range of sleep research covering from basic biology to pharmaceutical science and further to experimental medicine will be needed. Although the above research objectives are extremely ambitious, with wide scopes and it will take considerable time to be achieved, significant progress has been made toward attaining the objectives, as described in text 2-1.

Organization/operation

The basic concept of the organization and the operation of IIIS involves creating a new style of research center at Tsukuba by learning from the merits and virtues in the organization of "departments" in major U.S. universities. The strong leadership of the "Department Head" would be the first feature, thus we assigned similar authorities to the Director Masashi Yanagisawa, who had served as a professor/principal investigator for 20 years at University of Texas Southwestern Medical Center (UTSW). Other characteristics of this "department-style" operation include; (1) Flexible and timely appointment of principal investigators (PIs) at the discretion of Director, (2) Appointment of independent PIs regardless of their age and career stages with a necessary startup package, (3) A flexible and dynamic allocation of the floor space for each laboratory considering the laboratory's scale of funding, number of personnel and facility requirements, and (4) Sharing of large facilities and capital equipment among laboratories. Indeed, all of these characteristics are perfectly realized in the organization and operation of IIIS.

Likewise, the basic concept of the administration department is to construct a support organization that allows researchers to focus on their research without being hampered by many miscellaneous tasks. We recruited Ph.D. holders with experiences of drug discovery and/or liaison officer in industries for three key positions in the administration. This pre-empts the University Research Administrator (URA) system, which has been focused on recently by the Ministry of Education, Culture, Sports, Science and Technology (MEXT), and it constitutes one of the IIIS characteristics.

Research environment

Thanks to the MEXT subsidy for facility improvement and additional funding by the university, the IIIS Building (6-stories with 8,000 m^2 of floor space) was constructed in 2015. The IIIS Building serves as a globally unrivaled venue for conducting interdisciplinary sleep research under one roof, covering 3 research fields of i) basic biology such as molecular genetics and neuroscience, ii) pharmaceutical science including pharmacology and medicinal chemistry, and iii) experimental medicine like clinical sleep physiology and translational research. The 5/6th floors are dedicated to the Animal Facility exclusive to IIIS, cementing its role as a leading global research resource and far surpassing the original plan.

2. Center's Research Activities

Outlines

The perspective view of our research activities has been given in section 1. Our approaches to achieve

those objectives and specific subjects/themes set forth by each lab are as follows.

1) Elucidation of the fundamental mechanisms of sleep/wake regulation

Our current knowledge on sleep/wake regulation is actually quite limited when judged under the rigorous standards of today's neuroscience. We dissect neuronal and molecular mechanisms of sleep regulation to elucidate operating principles of neural networks regulating sleep/wake. At the same time, we use a completely unbiased genetic approach in order to identify new and unexpected genes that are importantly involved in the regulation of sleep/wake.

[1] Forward genetics to explore genes controlling sleep/wake regulation (Yanagisawa/Funato Lab)
[2] Reciprocal interaction between the extended amygdala and arousal systems (Sakurai/Sakaguchi Lab)
[3] The causal role of the mesolimbic brain system in the control of sleep and wakefulness (Lazarus Lab)
[4] Cortical neural networks in slow-wave sleep (Greene/Vogt Lab)
[5] Interaction of sleep, memory and adult neurogenesis (Sakurai/Sakaguchi Lab)
[6] Elucidation of the function of REM sleep (Hayashi Lab)

2) Elucidation of molecular pathogenesis of sleep disorders and related diseases

Irregular sleep/wake cycle and insomnia are risk factors for metabolic syndrome as well as for anxiety disorders. However, the mechanism for the link is unknown. Using mouse models, the possible molecular links between sleep/wake, mood regulation and metabolic control are studied.

[7] Establishment of a REM sleep behavior disorder (RBD) model and translational research using the orexin antagonist (Yanagisawa/Funato Lab)
[8] Forward genetics to understand the molecular basis of fear and fear/anxiety disorders (Liu Lab)

3) Development of treatments for sleep disorders

We develop new drug-candidate compounds modulating sleep/wake that are different from existing sleep-inducing agents or psychostimulants in their mechanism of action. We also develop methods for prevention and early intervention of sleep disorders and related diseases. This includes behavioral modifications to specific aspects of lifestyle, such as sleep, diet, exercise, and stress-coping. Those new drugs and intervention programs are not only effective for sleep disorders, but also for mood disorders and metabolic diseases. We utilize such associations in order to elucidate the molecular mechanisms behind the phenomena.

[9] Design and synthesis of orexin agonists (Nagase and Yanagisawa/Funato Lab)
[10] Identification of somnogenic natural compounds and elucidation of their mechanisms (Urade Lab)

Collaborative studies

Along with a close collaboration with UTSW, we set new satellite institutions such as the UC Berkeley (from 2014) and Kyoto University (from 2015) to reinforce research cooperation. Moreover, a joint research on the impact of the closed environment of space on sleep was launched in collaboration with JAXA. In addition to Akita University and RIKEN Bioresource Center, we newly expanded collaborations with; RIKEN Brain Science Institute to study on the physiological significance of sleep toward developing therapy for mental disorders; the Center for Genomic Medicine, Kyoto University for genome epidemiological studies of genetic/environmental factors related to human sleep.

Acquisition of research funds

The acquisition of competitive research grants is steadily growing. Research funds from special joint research projects, such as the ones concluded with Ibaraki Prefecture and with multiple companies including a global pharmaceutical company, are also growing constantly. Outcomes of those joint research projects will be practically exploited and returned to the society in the future. In particular, we work with pharmaceutical companies to focus on drug discovery research as well as clinical research of human sleep in cooperation with sleep-related companies/research institutes, covering a range of translational research from basic to clinical. Those research have already resulted in and provided promising applications and implementations with about ten patents (under the review processes).

3. Interdisciplinary Research Activities

To achieve the three IIIS research objectives listed in section 1, basic biology, experimental medicine and pharmaceutical science are fused to promote multifaceted sleep research. IIIS was established as a research organization under the direct supervision of the President of University of Tsukuba, and a system to facilitate the implementation of interdisciplinary research has been organized. The Director is a medical doctor with a strong background in pharmacology, also a renowned pioneer of sleep neuroscience, demonstrating the powerful leadership towards creating a novel research field, "sleep science".

For the core and satellite groups of IIIS, we assigned PIs with sufficient experiences and achievements in areas of pharmaceutical science and experimental medicine as well as PIs active in basic biology and

encouraged to link together. The Administrative Director, who experienced the director of research center of a pharmaceutical company, was also assigned to supervise the coordination of translational research. Many PIs of IIIS are enrolled with medical qualifications, and as well many other staff members have experienced drug discovery researches in pharmaceutical companies. Those personnel set-up also boosts establishing fusion research in three areas in a bottom-up structure. The new research building is the open-office format, with a structure to facilitate interactions and communication among members within and outside laboratories. In addition, various meetings and seminars are also arranged to create opportunities where members from different laboratories interact with.

To enhance the research system toward the creation of a fusion area, we commenced the joint research with a global pharmaceutical company and will appoint a satellite PI from the company. Some research areas in pharmaceutical science, such as pharmacokinetics, pharmacodynamics, toxicology, etc. are implemented as part of the joint research, because those are difficult to conduct at universities. Some other collaborations are also underway with universities and national institutes including JAXA.

4. International Research Environment

Overseas core PIs actively participate in the management of IIIS. Liu and Greene in particular, besides conducting researches at the satellite institute (UTSW), have also established their own laboratories in the IIIS core group, and stayed for one to three months a year to engage in research activities in Japan. They also actively participated in the monthly PI meeting (via Skype when they are in US), international symposia and MEXT/JSPS site visits.

For the recruitment of overseas young researchers, we have employed 10 brilliant scientists through the continuous efforts in international recruitment or networks of PIs. We also actively engaged in recruiting at the international symposia held by IIIS and the other conferences to acquire a new female satellite PI.

The administrative department of IIIS has been supporting foreign researchers in various procedures, e.g. providing information, document preparations and visa application, in cooperation with other branches in the university (Global Commons, International Exchange Support Office) and the Japan International Science and Technology Exchange Center (JISTEC). Many foreign researchers use accommodations operated by JISTEC. By assigning staff members (both admin staff members and secretaries) with high English proficiency, many forms (for applications, recruitment, personal affairs and general affairs) are translated in English, achieving a foreigner-friendly atmosphere with sufficient attentive supports.

5. Implementing Organizational Reforms

By positioning IIIS as an independent department of the university, extensive independent operations are secured, including personnel affairs, environmental maintenance and budget implementation. The important matters were determined by the top-down responsibility of the Director. To ensure the intention of the Director can be swiftly reflected, ongoing efforts are made to improve the relevant rules, such as detailed departmental regulations. Furthermore, some efforts for system reform were made as follows;

- Led by the administrative department, we hold a PI meeting on the regular basis to decide important matters concerning the institute where PIs and Director mutually and freely exchange their ideas.
- IIIS Personnel Committee engaging in dedicated discussion allowed IIIS to expedite judgment and appointments compared to a conventional personnel committee in the university.
- Ensuring English as the official language, ~60% of administrative staff members are bilingual.
- Despite the external advisory board has been considered, we have not proceeded with the selection of members for the board due to budgetary constraints (because the annual subsidy is unexpectedly less than the original plan). We are currently considering a novel system to evaluate researchers by the index based on research achievements (e.g. publications, external funds acquired and achievements in cooperation with industry, government and schools).
- To enable Yanagisawa to concurrently hold positions in the University of Tsukuba and UTSW, President Nagata took the initiative to newly introduce a Joint Appointment System to the university in March 2014.
- The animal facility on the 5/6th floors of the new research building (ARC Satellite) is operated in cooperation with the Laboratory Animal Resource Center (ARC) of the University of Tsukuba. This advanced facility plays a pilot role in animal breeding techniques among facilities in our university.

6. Future Vistas

The crucial issues to achieve the initial concept of IIIS are being tackled as follows:

1. Improving and maintaining facilities and equipment to achieve a “World Premier International” standard

- Expanding the breeding capacity of the animal facility (ARC Satellite)
- Establishing a clinical sleep laboratory
- Implementation of the future expansion space

2. Further developing strength of IIIS such as capabilities of neuroscience
- Transfer/relocation of the staff members and equipment of Sakurai laboratory of Kanazawa University

3. Filling gaps in research capabilities in pharmacokinetics, toxicology, etc.
- Pursuing research collaboration with pharmaceutical companies in drug discovery research involving therapeutic agents for sleep disorder other than orexin agonists

4. Strengthening capabilities in clinical sleep physiology, human molecular genetics, etc.
- Promoting translational/clinical research in collaboration with satellites and other research institutes

To secure sufficient research funds, we will further make continuous efforts to obtain competitive research grants, and promote joint research with companies focusing on the development of sleep-promoting products, including pharmaceutical companies in future.

We consider it crucial for IIIS to remain "the leading global research center", which will require establishing a system where license revenues for research results can be directly reinvested in IIIS. In the initial applications, the University of Tsukuba has committed to maintain IIIS as a permanent research institute even after the period of WPI program expires. As part of specific measures, discussions about the tenure-truck system for PIs were held between the Vice Presidents for Research and Personnel Affairs of University of Tsukuba.

7. Others

One of the important progresses in the management of IIIS in FY2015 was to invite Sakurai to Tsukuba to set him as a Vice Director. Aiming to enlarge the research capability of the IIIS in the field of neuroscience, the operational management system and the leadership structure for young researchers, we decided to physically install him from April 1, 2016. Vice Director Sakurai will be employed as a succession employee, concurrently works for the Faculty of Medicine and IIIS.

By obtaining financial contributions from the Seven Dreamers Laboratories Inc., Satoh Lab, established by the implementation of a special joint research project with Ibaraki Prefecture, will be expanded as an endowed laboratory of clinical sleep medicine. This is only the second case in 17 years in which those department systems have been established in the University of Tsukuba.

8. Center's Response to Results of FY2015 Follow-up (including Site Visit Results)

Q: Results of forward genetics studies and genes identified (*Sleepy 1/ 2*, *Dreamless*) should be published before the interim evaluation, and the strategy to analyze the functional roles of those genes at molecular level (e.g. signal transduction, gene expression profiling, etc.) should be stated.

A: The first pioneering article for the discovery of *Sleepy* and *Dreamless* gene is now ready for submission and we are currently waiting for the reviews. Multiple articles depicting functional aspects of those genes and protein products are under preparation and will be submitted in the near future. Details are described in the texts.

Q: The reduction in external funding is a significant concern.

A: The amount of external competitive funds in FY2015 almost doubled compared to FY2014. Although it remains insufficient, we continuously make every effort to acquire more competitive funds and we expect further improvements toward FY2016.

Q: How does IIIS translate results obtained from basic research using animals into human studies?

A: We fully understand the importance and consider it crucial to translate the results of basic studies to the clinical studies, and thus we have initiated multiple programs with external institutes/companies. Details are described in the main text.

Q: Employment of female PIs in the core group should be seriously considered.

A: We have been seeking a good candidate for PI positions by posting ads for female-oriented recruitment since Oct 2014. Although not yet succeeded, we will continue recruiting efforts. We are also concurrently considering the development of female researchers to appoint as young PIs.

Q: Given increasing number of students, an appropriate mentoring system is required for IIIS.

A: We understand the importance of the mentoring system: so far PIs become mentors after receiving certifications as instructors from graduate schools of the University of Tsukuba, and non-PI faculties assist them when necessary, which is a typical approach in the University of Tsukuba.

World Premier International Research Center Initiative (WPI)
Self-Evaluation Report for Interim Evaluation

Host Institution	University of Tsukuba	Host Institution Head	Kyosuke Nagata
Research Center	International Institute for Integrative Sleep Medicine	Center Director	Masashi Yanagisawa

Common Instructions:
* Please prepare this report based on the current (31 March 2016) situation of your WPI center.
* As a rule, keep the length of your report within the specified number of pages. (The attached forms are in addition to this page count.)
* Use yen (¥) when writing monetary amounts in the report. If an exchange rate is used to calculate the yen amount, give the rate.

1. Summary of State of WPI Center Project Progress (write within 2 pages including this page)

Describe the center's identity and the achievement status of its initially stated goals.

- On the sheets in Appendix 1~5, list the Principle Investigators, and enter the number of center personnel, a chart of the center's management system, a campus map showing the center's locations on the campus, and project funding.

Research activity

The new interdisciplinary domain we aim to create is "sleep science." It is an interdisciplinary branch of the biomedical field that focuses on sleep, while it is supported by three main pillars: basic biology, pharmaceutical science and experimental medicine.

Sleep is a behavior that everyone experiences daily and it takes up as much as one third of one's entire lifetime. However, the very fundamental mechanisms of sleep and its *raison d'être* remain still unknown today. While sleep has been a black box stubbornly resisting scientists' challenges, its medical and social importance is very clear. Healthy sleep is necessary for maintaining our mind and body fitness; lack of sound sleep not only causes a reduction in higher brain functions including memory and decision making, but also increases the risk of mood disorders such as depression as well as metabolic syndrome, etc. In developed countries, the prevalence rate of sleep disorders is around 15%, with the lifetime prevalence more than 30%. The underlying factors behind this problem include an increase of the elderly population and the increasingly nocturnal lifestyle of today's societies. Shift workers now account for 10% of the working population, and a large part of them, 20-30% are suffered from sleep disorders. The deficiencies in healthy sleep cause significant social losses, and are linked to decrease in working efficiency and increase in accidents due to excessive sleepiness, and increased prevalence of mood disorders and metabolic syndromes, and even increased suicide deaths. Domestic economic loss caused by sleep disorders is estimated as 3.5 trillion yen/year, hence it is the urgent need to solve sleep-related issues.

We set out our **major research objectives** to solve the issues of sleep disorders as follows.

1) To elucidate the fundamental mechanisms of sleep/wake regulation
2) To elucidate molecular pathogenesis of sleep disorders and related diseases
3) To develop treatments for sleep disorders

To achieve these objectives, there is a need for wide-ranging sleep research, covering a scope from basic biology such as neuroscience to pharmaceutical science and further to experimental medicine. This is "sleep science" which embodies the research activities of this institute as its identity. Although the above research objectives are extremely ambitious, with wide scopes and it will take considerable time to be achieved, significant progress has been reached toward attaining the respective objectives, as in 2-1.

A major challenge to establish "sleep science" is, as advised by the working group of the site visit, the implementation of translational research with which we translate achievements of basic biology and pharmaceutical science to experimental medicine and/or clinical research. To implement the translational research, we take two measures; expanding the clinical group in IIIS and the research alliances with outside groups. Since resources to enlarge the inside group is rather limited, major efforts have been dedicated to increase and expand collaboration programs with outside groups, as described in 2-2.

Organization/operation

The basic concept of the organization and the operation of IIIS involves creating a new style of research center at Tsukuba by learning from the merits and virtues in the organization of "departments" in major U.S. universities. We could implement the WPI's mission and mandate aiming at "system reform" by selectively learning from the merits of department organizations in the U.S. academia. The strong leadership of the "Department Head" of a U.S. university would be the first feature we should pick up,

and we thus assigned similar authority to the Center Director, Masashi Yanagisawa, who had served as a professor/principal investigator for 20 years at University of Texas Southwestern Medical Center (UTSW), one of the best biomedical campuses in the U.S. Other characteristics of this "department-style" organizational operation we would like to adopt include:

- Flexible and timely appointment of principal investigators (PIs) at the discretion of the Department Head within the budget limitation,
- Appointment of independent principal investigators regardless of their age and career stage with a necessary startup package,
- A flexible and dynamic allocation of the floor space for each laboratory considering the laboratory's scale of funding, number of personnel and facility requirements, and
- Sharing of large facilities and capital equipment among laboratories.

Indeed, all of these characteristics are perfectly realized in the organization and operation of IIIS. There are four young PIs in the core group of IIIS. One of them has been PI since his appointment as an assistant professor in 2012 and promoted to an associate professor recently. The labs and offices in the new research building are designed as open labs and open offices, respectively, which enable the flexible and dynamic allocation of the floor space. The basic concept of the organization and the operation surely motivates young scientists and contributes to free interaction and open communication throughout IIIS, and hence vitalizes the whole research activities of IIIS.

Likewise, concerning the administration department, it is the basic concept to construct a support organization that allows researchers to focus on their research without being hampered by many administrative and miscellaneous tasks. To this end, we recruited Ph.D. scientists that have experiences of drug discovery and/or liaison officer in industries for three key positions in the administration, *i.e.*, Administrative Director, the leader of Research Planning, and the leader of Outreach Activity, to act as an interface between researchers (the Center Director and PIs) and the administrative staffs without scientific background in the administration as well as the university headquarters. This pre-empts the University Research Administrator (URA) system, the development of which has been focused on recently by the Ministry of Education, Culture, Sports, Science and Technology (MEXT) and which constitutes one of the IIIS characteristics.

Research environment

In the original plan we proposed in the application for the WPI program, IIIS were supposed to be provided with 5,000 m^2 of floor space by continuing to use the floors on the Health and Medical Science Innovation Laboratories and by occupying all floors on the renovated E Building of the University Tsukuba Hospital. Thanks to the MEXT subsidy for facility improvement and additional funding by the university, the IIIS Building (6-stories with 8,000 m^2 of floor space) was constructed at the north corner of the Hospital Area, laying side-by-side buildings of biomedical research in the Medical School. The IIIS Building serves as a globally unrivaled venue for conducting interdisciplinary sleep research under one roof, covering 3 research fields of i) basic biology such as molecular genetics and neuroscience, ii) pharmaceutical science including pharmacology and medicinal chemistry, and iii) experimental medicine like clinical sleep physiology and translational research.

The increased floor space of 3,000 m^2 is dedicated to the vivarium exclusive to IIIS on the 5th and 6th floors. The breeding area on 6th floor accepts up to 6,000 IVC, and the experimental area on 5th floor accommodates 7 sleep recording labs and 6 behavior labs, cementing its role as a leading global research resource and far surpassing the original plan.

2. Center's Research Activities (within 8 pages)

2-1. Research results to date

Provide an overall picture of the Center's research activities and select 5~10 representative results achieved during the period from 2012 through March 2016. Number the results [1] to [10] and provide a description of each.

・In Appendix 2-1, list the papers underscoring each research achievement and provide a description of each of their significance.

The perspective view of our research activities has been given above (1. Summary of State of WPI Center Project Progress). The research objectives that we seek to achieve through the research activities of sleep science are: 1) Elucidation of the fundamental mechanisms of sleep/wake regulation, 2) Elucidation of molecular pathogenesis of sleep disorders and related diseases, and 3) Development of treatments for sleep disorders. Our approaches to achieve these objectives and specific subjects/themes set forth by each lab to realize the approaches are as follows.

1) Elucidation of the fundamental mechanisms of sleep/wake regulation

Our current knowledge on sleep/wake regulation is actually quite limited when judged under the rigorous standards of today's neuroscience. Among the limited tidbits is the notion that the lateral hypothalamic orexin neurons and the wake-active monoaminergic and cholinergic neurons of the classical ascending activation system, together with the sleep-active GABAergic neurons of the preoptic hypothalamus, likely constitute important parts of the executive circuitry for sleep/wake switching. Orexin neurons are clearly important for the stability of the switch. We know that the sleep-inducing substance adenosine (which is blocked by caffeine) is importantly involved in the regulation of the "depths" of non-REM sleep. We know that these executive systems are powerfully governed by the circadian clock in the suprachiasmatic nucleus and by the presumed "sleep homeostat" somewhere in the brain. Overall, our current level of understanding is rudimentary at best.

We conduct precise neurophysiological analyses of these known components. We dissect neuronal and molecular mechanisms of sleep regulation to elucidate operating principles of neural networks regulating sleep/wake. At the same time, we use a completely unbiased genetic approach in order to identify new and unexpected genes that are importantly involved in the regulation of sleep/wake.

[1] Forward genetics to explore genes controlling sleep/wake regulation (Yanagisawa/Funato Lab)
[2] Reciprocal interaction between the extended amygdala and arousal systems (Sakurai/Sakaguchi Lab)
[3] The causal role of the mesolimbic brain system in the control of sleep and wakefulness (Lazarus Lab)
[4] Cortical neural networks in slow-wave sleep (Greene/Vogt Lab)
[5] Interaction of sleep, memory and adult neurogenesis (Sakurai/Sakaguchi Lab)
[6] Elucidation of the function of REM sleep (Hayashi Lab)

2) Elucidation of molecular pathogenesis of sleep disorders and related diseases

Irregular sleep/wake cycle and insomnia are a risk factor for metabolic syndrome as well as for anxiety disorders. However, the mechanism for the link is unknown. Using genetically engineered mouse models, the possible molecular links between sleep/wake, mood regulation, and metabolic control will be studied.

[7] Establishment of a REM sleep behavior disorder (RBD) model and translational research using the orexin antagonist (Yanagisawa/Funato Lab)
[8] Forward genetics to understand the molecular basis of fear and fear/anxiety disorders (Liu Lab)

3) Development of treatments for sleep disorders

We will develop new drug-candidate compounds modulating sleep/wake that are different from existing sleep-inducing agents or psychostimulants in their mechanism of action. We will also develop methods for prevention and early intervention of sleep disorders and related diseases. This includes behavioral modifications to specific aspects of lifestyle, such as sleep, diet, exercise, and stress-coping. It is likely that these new drugs and intervention programs are not only effective for sleep disorders, but also for mood disorders and metabolic diseases. We will utilize such associations in order to elucidate the molecular mechanisms behind the association.

[9] Design and synthesis of orexin agonists (Nagase and Yanagisawa/Funato Lab)
[10] Identification of somnogenic natural compounds and elucidation of their mechanisms (Urade Lab)

Research results to date of each subject/theme are as follows;

[1] Forward genetics to explore genes controlling sleep/wake regulation (Yanagisawa/ Funato Lab)

As many of IIIS labs successfully utilize optogenetic and chemogenetic approach, a recent advance in directly manipulating the activity of specific neural circuitries has uncovered neural circuitries regulating sleep/wakefulness states. However, the molecular and cellular mechanism that determines the propensity of switching between wakefulness, NREM sleep and REM sleep remains unknown. To tackle this issue, we have conducted EEG/EMG-based screening of more than 8,000 randomly mutagenized mice, which led to identify three gene mutations resulting in sleep/wakefulness abnormalities (Funato *et al.*, *Nature* submitted; Fujiyama *et al.*, in prep; Kim *et al.*, in prep). We found that *Sleepy* mutant mice have a point mutation in the *Sik3* protein kinase gene at the splice donor site after exon13 and the complete skip of exon13 (Figure 1). We then succeeded in showing the causal relationship of the *Sik3* mutation by introducing gene modifications which results in the skip of exon13. SIK3 is a member of AMP-activated protein kinase (AMPK) family and is broadly expressed in both excitatory and inhibitory neurons of the forebrain and brain stem (Komiya *et al.*, in prep). The exon13 has a phosphorylation site which is well conserved among the animal kingdom, suggesting the conserved role of *Sik3* orthologues in sleep-like behaviors. In collaboration with Yu Hayashi of IIIS and Dr. Kazuhiko Kume, Nagoya City

University, we showed that the modification of *Sik3* orthologues altered the amount of sleep-like behaviors of roundworms and flies in a direction that is consistent with Sleepy mutant mice (Funato *et al.*, *Nature* submitted). Furthermore, we establish FLAG-SIK3 knock-in mice using CRISPR technology, which enables us to examine the phosphorylation status of SIK3 protein *in vivo*. In parallel, quantitative phosphoproteomic analysis of *Sleepy* mutant brains has been examined (Wang, Liu *et al.*, in prep). As there has been a total lack of knowledge about the intracellular signaling that regulates sleep/wakefulness, the identification of SIK3 protein will open new frontiers in sleep research.

As for REM sleep, we found a single amino acid substitution of the leak cation channel NALCN in *Dreamless* mutant mice (Figure 1) which show drastic reduction in the total amount and mean episode duration of REM sleep (Funato *et al.*, *Nature* submitted; Fujiyama *et al.*, in prep). CRISPR-based *Nalcn* mutant mice recapitulated the REM sleep abnormality. Mutant NALCN-transfected cells show larger ionic conductance than wild type NALCN-transfected cells, implying that NALCN may regulate cellular excitability of the brain stem neurons responsible for the switching out of REMS, which have been identified by Yu Hayashi, IIIS.

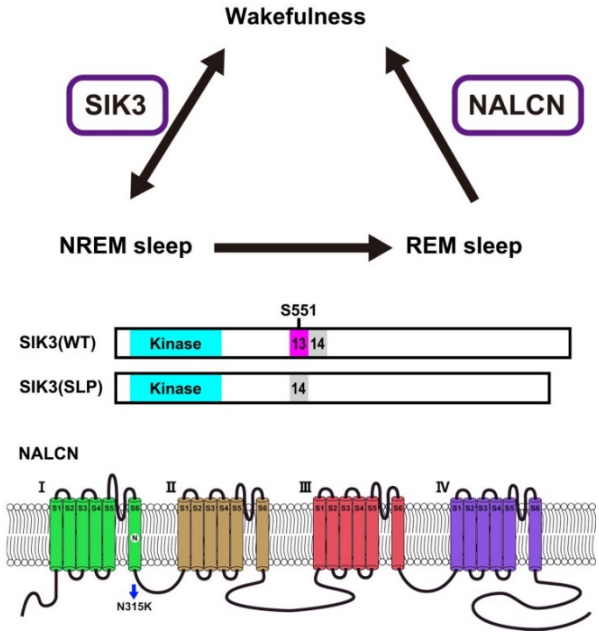


Figure 1: A forward genetic study identified mutations in *Sik3* and *Nalcn* genes of *Sleepy* mutant and *Dreamless* mutant mice, respectively. The splicing mutation in *Sik3* gene results in skipping of an exon13-encoded region of the SIK3 protein kinase. A point mutation in the *Nalcn* gene causes an amino acid substitution in the segment 6 of the domain I of the NALCN.

Importantly, *Sleepy* and *Dreamless* mutant mice also exhibit altered memory, mood and energy metabolism (Honda *et al.*, in prep). We have been working on to establish *Sik3*- and *Nalcn*-gene modified mice which can be regulated by Cre-lox system. These mice will enable us to identify neural groups responsible to each of the multiple behavioral and metabolic phenotypes.

[2] Reciprocal interaction between the extended amygdala and arousal systems (Sakurai/Sakaguchi Lab)

(Papers underscoring the research achievement above and their brief account: [Appendix 2-1] 1-3)

Orexin neurons are activated in response to emotionally salient cues (1). To reveal the mechanisms by which orexin neurons are regulated by salient cues and/or contexts, we identified neurons that make monosynaptic inputs to orexin neurons using a recombinant rabies virus-mediated trans-synaptic retrograde tracing in mice. We identified positive cells in the many brain regions implicated in emotion, reward system, and sleep. By combination of the cell type-specific tracing the relationship between input and output (cTRIO) analysis and anterograde tracing we found many GABAergic neurons in the POA, including the VLPO, which send projections to orexin neurons receive monosynaptic projections by neurons in the central nucleus of the amygdala (CeA) and BST (Saito *et al.*, unpublished). Pharmaco- or optogenetic activation of BST GABAergic neurons or their axon terminals in the POA during NREM sleep made immediate transition from NREM sleep to wakefulness in a phase-locked manner, while stimulation during REM sleep did not show any effects (Kodani *et al.*, unpublished). We also examined the function of orexinergic projections in the regulation of emotion-related behavior. We previously found that targeted restoration of orexin receptor expression in noradrenergic neurons of the locus coeruleus (LC) and in serotonergic neurons of the dorsal raphe (DR) in OX1R-/-;OX2R-/- mice, which display a severe narcoleptic phenotype, differentially inhibited fragmentation of wakefulness and cataplexy, respectively (2). We further found that optogenetic excitation of DR-5HT→lateral amygdala (LA) pathway almost completely inhibited cataplexy, which was induced by chocolate feeding in the mice (Hasegawa *et al.* unpublished). We also examined roles of the orexin neurons→LC-NA neurons→lateral amygdala (LA) pathway in the fear-related behavioral responses (3). After fear conditioning in the particular context, optogenetic stimulation of orexinergic fibers in the LC, or LC-NA fibers in the LA induced an apparent freezing behavior even in the in alternative context which did not induce freezing when the stimulation was not applied (Soya *et al.*, unpublished). Pharmacogenetic or optogenetic inhibition of LC-NA neurons reduced the freezing in the fearful context. These results suggest that orexin neurons activate the

amygdala projecting-LC-NA neurons to modulate fear-related behavior. These observations suggest that the extended amygdala regulates orexin neurons and monoaminergic systems, and arousal systems in turn influences amygdala function to modulate emotion-related behavior as well as arousal.

[3] The causal role of the mesolimbic brain system in the control of sleep and wakefulness (Lazarus Lab) (Papers underscoring the research achievement above and their brief account: [Appendix 2-1] 4-6)

Sleep control is ascribed to a two-process model, a widely accepted concept that posits homeostatic drive and a circadian process as major sleep-regulating factors. Sleep/wake behaviour, however, is also influenced by cognitive and emotional factors, which are not accounted for by the two-process model (4, 5), and thus the brain mechanisms involved in controlling sleep remain an open question. The arousal effect of caffeine depends on adenosine A_{2A} receptors ($A_{2A}R$) on neurons in the nucleus accumbens (6), which is a major component of the mesolimbic brain system. $A_{2A}R$-positive NAc inhibitory medium spiny neurons also express dopamine D2 receptors (D2R) and thus, are involved in the dopaminergic control of motor function and motivational behavior. However, their role in the regulation of sleep was not known. The lab found that chemogenetic and optogenetic stimulation of the indirect pathway in the NAc induced robust slow-wave sleep, whereas slow-wave activity was not affected.

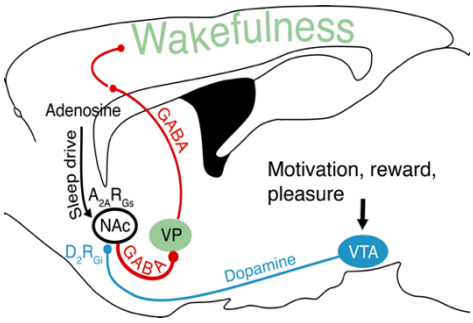


Figure 2: Novel brain circuit for sleep control by motivated behavior. In the absence of cognitive and emotional stimuli, NAc neurons induce sleep by depressing wake-promoting VP neurons in the basal forebrain.

As the NAc contains two anatomically and functionally different components, shell and core, they examined whether these structures have distinct roles on sleep regulation and found that the optogenetic stimulation of the indirect pathway in the NAc core promotes slow-wave sleep. Although the NAc core innervates many arousal-related brain areas, including the lateral hypothalamus which produces orexin, the tuberomammillary nucleus which produces histamine, and the ventral tegmental area which produces dopamine, the lab found that selective activation of neuronal projections from the NAc core to ventral pallidal (VP) neurons in the basal forebrain induced slow-wave sleep. Their findings reveal a prominent contribution of this indirect pathway to sleep control (Oishi *et al., Science*, submitted). This novel brain circuit may explain the tendency to fall asleep in the absence of motivating stimuli, i.e., when bored. The tonic sleep drive by neurons in the NAc core may be inhibited by ongoing cognitive and emotional stimuli, but in the absence of such stimuli, i.e. under low dopamine conditions, may allow the brain to fall asleep by depressing firing of arousal circuits in the basal forebrain (Figure 2). This interpretation is consistent with other work in the lab showing that chemogenetic activation of midbrain dopaminergic neurons strongly consolidates wakefulness through D2R, but not dopamine D1 receptors, suggesting a D2R-dependent arousal system in the midbrain (Oishi *et al.*, *eLife*, in prep).

[4] Cortical neural networks in slow-wave sleep (Greene/Vogt Lab)

(Papers underscoring the research achievement above and their brief account: [Appendix 2-1] 7-9)

Waking causes the accumulation of sleep need and controls sleep intensity in subsequent sleep phases. Longer periods of waking lead to deeper sleep with increased slow wave activity (SWA). Our recent publication provides evidence for an adenosine-mediated regulation of sleep in response to waking (*i.e.*, homeostatic sleep need), requiring activation of neuronal adenosine A1 receptors and controlled by glial adenosine kinase (Figure 3) (7). Thus slow wave activity is a critical indicator of both sleep need buildup and resolution consistent with a role in sleep function. We are now implementing and refining tools for

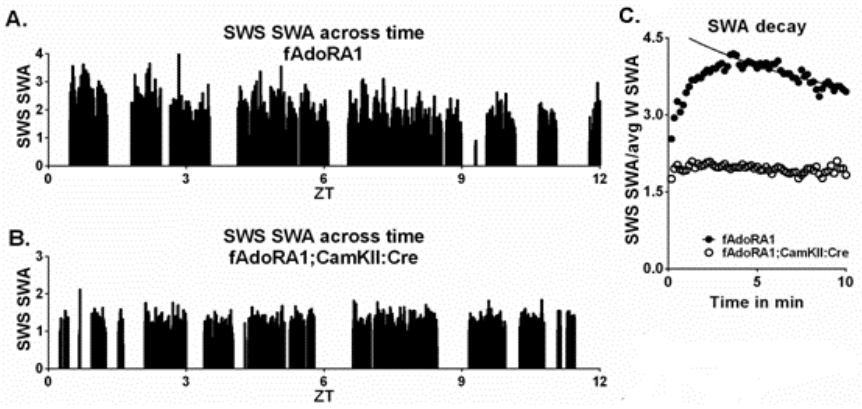


Figure 3: Homeostatic reaction of slow wave activity (SWS SWA) in mice and its dependence on neuronal adenosine receptor function. SWA is initially high and then decays during sleep phases (A, C black dots), indicating buildup and resolution of sleep need. This process is perturbed in mice lacking AdoRA1 in excitatory cortical neurons (B, C open circles).

the assessment of cortical local circuit function in the generation and changes in SWA. This includes *in-vivo* multi-electrode recordings and functional imaging, using sensors for both calcium concentration and transmembrane voltage (8, 9).

We aim to understand the functional transition from wake to slow wave sleep in excitatory- inhibitory cortical networks and their homeostatic regulation. In addition we are analyzing frontal-cortical (the area with the highest amplitude SWA) differentially expressed genes across sleep conditions. It is plausible that Adora1-dependent increase of Ca^{2+} during SWS-SWA is a trigger for our preliminary finding that more than 2,900 genes have altered expression across sleep states. This can be directly tested with our floxed Adora1 mutants.

[5] Interaction of sleep, memory and adult neurogenesis (Sakurai/Sakaguchi Lab)

(Papers underscoring the research achievement above and their brief account: [Appendix 2-1] 10-16)

Human adult neurogenesis takes place almost exclusively in the hippocampus. These adult-born neurons could be used to regenerate damaged brain tissue (10, 11). For this, it is essential to know how they integrate into existing neuronal circuits (12).

We were the first to demonstrate that adult-born neurons are functionally incorporated into memory circuits (13, 14). Recently, we reported that there is a critical time period in fear memory formation (15). By sleep stage-specific manipulation of adult-born neuron during this time period using optogenetics (16), we discovered an important function of adult-born neurons in memory formation (manuscript in prep).

Currently, we are trying live-imaging of the activity of adult-born neurons during sleep by combining activity-specific synaptic tagging technology and original microendocope in collaboration with Dr. Akiko Takagi at Univ. Gunma, Japan and Dr. Alcino Silva at UCLA, U.S. Moreover, we have started to examine a transgenic primate model of a neurodegenerative disease in collaboration with Dr. Hideyuki Okano in Keio Univ., Japan. These studies could potentially lead to establish a new therapeutic method using adult-born neurons for patients with neurodegenerative diseases.

[6] Elucidation of the function of REM sleep (Hayashi Lab)

(Papers underscoring the research achievement above and their brief account: [Appendix 2-1] 17)

The function of REM sleep is one of the largest mysteries in neuroscience. During REM sleep, both cortical neural activity and blood flow increase. To address the roles of REM sleep, we identified neurons in the brainstem that either inhibit REM sleep (17) or promote REM sleep (Kashiwagi *et al.*, unpublished), and established transgenic mice in which REM sleep can be artificially suppressed or increased (Figure 4). As a result, till now, we found that artificial suppression or induction of REM sleep attenuates or enhances slow wave activity (SWA) during subsequent NREM sleep, respectively. These results obtained from our novel mouse models suggest that REM sleep promotes slow wave activity during NREM sleep (17). Slow waves are known to promote memory consolidation and synaptic plasticity, and thus our study implicates that REM sleep is important for these events. To further elucidate the beneficial roles of REM sleep, we are now combining our mouse models with various neurological disease models and investigate whether we can improve the symptoms by either increasing or reducing REM sleep.

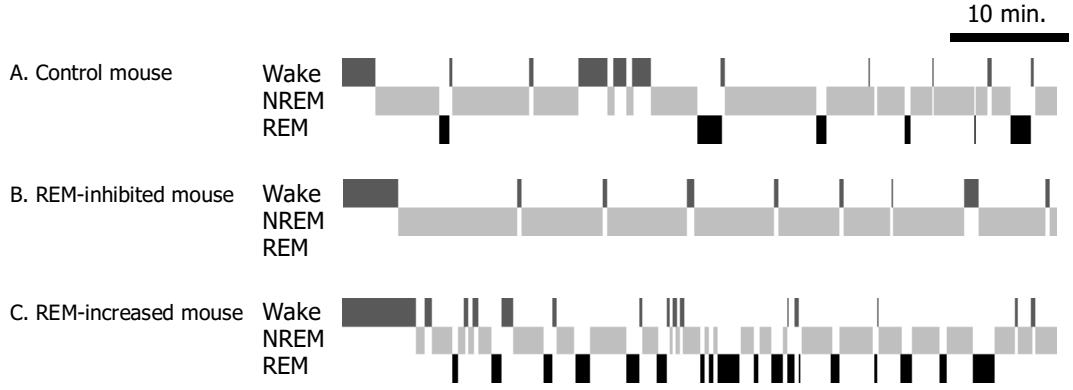


Figure 4:
Transgenic mice that enable REM sleep manipulation. A) During sleep, a wild-type mouse cycles between NREM sleep, REM sleep, and short wake; B) REM sleep inhibition by chemogenetic activation of brainstem REM-inhibiting neurons; C) REM sleep increase by chemogenetic activation of brainstem REM-inducing neurons.

[7] Establishment of a REM sleep behavior disorder (RBD) model and translational research using the orexin antagonist (Yanagisawa/Funato Lab)

Increasing attention has been paid to REM sleep behavior disorder (RBD) as a prodromal marker of Parkinson's disease and related neurodegenerative diseases. To develop a good animal model of RBD is crucial for understanding the pathophysiology of RBD and developing therapeutic intervention for it. Based on our hypothesis that disrupted glycinergic system underlies RBD symptoms, we have systematically examined Glra1-gene modified mice using Cre-loxP system. We then succeeded in developing RBD model mice ($Glra1^{flox/flox}$;$ChAT\text{-}Cre^{Cre/wt}$) which displayed gross body and limb movements including running, jerking and chewing during REM sleep. These RBD phenotypes were rescued by clonazepam that are routinely used to treat human RBD symptoms. Surprisingly, RBD behaviors of our RBD model mice were drastically suppressed by orexin receptor antagonists. Then, we immediately began collaborating with one of major sleep clinics to examine the clinical benefit of orexin receptor antagonists. From human study, we found 78% of RBD patients were treated effectively with the orexin receptor antagonists (Figure 5). Thus, our RBD model mice would serve as a platform of screening for a novel drug for treating RBD.

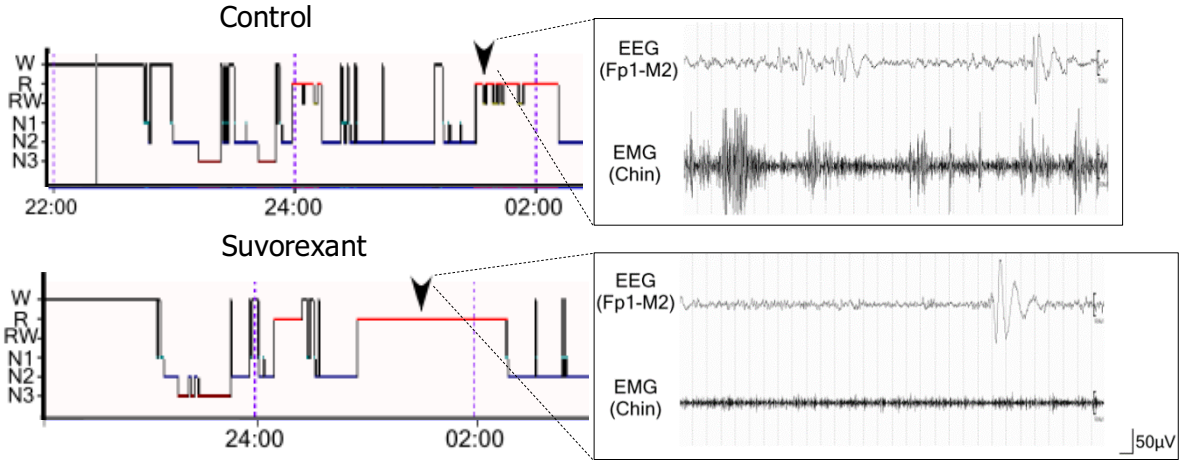


Figure 5: Orexin receptor antagonists (suvorexant) rescue the RBD phenotype in human RBD patients. In hypnograms for control (upper panel) and suvorexant (lower panel), arrow heads showed typical raw EMG and EEG traces. W; wake, R; REM sleep, RW; RBD, N1-N3; non-REM sleep stage1-3.

[8] Forward genetics to understand the molecular basis of fear and fear/anxiety disorders (Liu Lab)

Emotions define the essence of being human and are powerful drivers of behaviors. Fear is a basic emotion that enhances animal survival by triggering the fight, flight or freeze responses to perceived danger. And yet emotions are complex and difficult to quantify and study. Almost nothing is understood about emotions at the molecular level. We have been conducting a forward genetic (from phenotype to gene) screen, based on a novel predator odor-induced innate fear assay that we developed, to identify mutations that either mitigate or accentuate innate fear in mice. After screening 15,987 ENU-mutagenized mice in two (a dominant and a recessive) fear screens, we isolated multiple heritable "fearful" and "fearless" mutant pedigrees and identified four putative fear proteins. In particular, one fear protein-a calcium channel-has been characterized in detail, elucidating a novel sensory pathway for predator odor-induced innate fear behaviors (Wang *et al.*, in prep). This is the first forward genetic screen on emotions, which will allow us to identify core fear genes and understand the molecular circuitry of fear. Our studies may usher in a new era of molecular investigations into the fundamental principles of human emotions. Furthermore, hundreds of millions of people worldwide suffer from many fear/anxiety disorders, including post-traumatic stress disorder (PTSD), general anxiety disorder, general or specific phobia, and obsessive compulsive disorder (OCD). The molecular

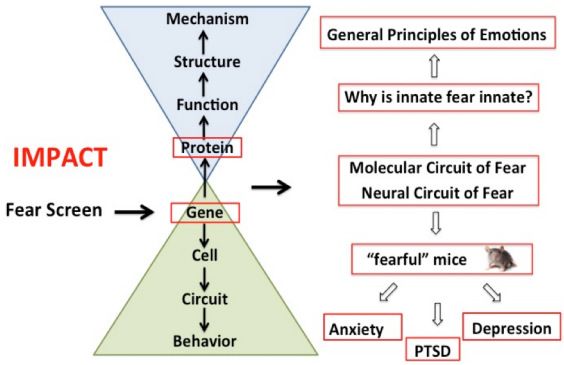


Figure 6: Strategy to elucidate molecular basis of fear and anxiety disorders

underpinnings of these brain diseases are unknown. Our studies will identify specific molecular targets and animal models to advance treatments for fear/anxiety disorders, promoting resilience of neurological health and optimizing human aptitude and performance (Figure 6).

[9] Design and synthesis of orexin agonists (Nagase and Yanagisawa/Funato Lab)

(Papers underscoring the research achievement above and their brief account: [Appendix 2-1] 18-19)

The small molecules showing agonist activity for orexin receptors, especially for OX2R, have been expected as a chemotherapeutic agent for narcolepsy. In 2012, Nagase's group discovered the potent OX2R selective agonists such as YN-1055 (EC_{50} = 23 nM) from > 1,500 synthetic samples, which were originally designed based on the structure of high throughput screening hit compounds in the University of Texas Southwestern Medical Center. While YN-1055 exhibited desirable activity *in vitro*, it was hard to solve in water. Therefore, we tried to modify the compound to improve the water solubility, leading to find a water-soluble OX2R agonist YNT-185 (EC_{50} = 28 nM; selectivity ratio to OX1R over 100 times). The ip (40 mg/Kg) and icv (260 nmol in 6 μL) administration of YNT-185 showed not only induction and duration of wake time in wild-type mice but also significant anti-narcoleptic effect in chocolate induced murine narcoleptic model (18, 19). These effects were not observed in orexin receptor KO mice. The physicochemical properties of YNT-185 were improved using Wager's CNS multi-parameter optimization (CNS MPO) method with predicted physicochemical values. The design and synthesis of > 1,000 compounds with the in silico evaluation led to identify YNT-1114 (EC_{50} = 31 nM) with desirable CNS MPO score of 3.5 (CNS MPO score >3.0 is desirable for CNS drug). Evaluation of its pharmacological effects *in vivo* is undergoing (Figure 7).

In the course of the above SAR studies, a dual agonist for OXRs and a selective agonist for OX1R (EC_{50} = ~900 nM) were identified, which would be good lead compounds to obtain the desired agonists with high affinity and selectivity for OX1R. Recently, we also found that nalfurafine, a kappa opioid agonist showed antagonistic activity for OX1R (IC_{50} = 415 nM) and was modified to lead higher affinity and selectivity for OX1R. Finally, the IC_{50} values were attained to be about 2 nM and selectivities were > 10,000 times to OX2R. These compounds are the best antagonists for OX1R in the world. We will also try to clarify the pharmacological effect of OX1R using the above agonist and antagonist selective for OX1R.

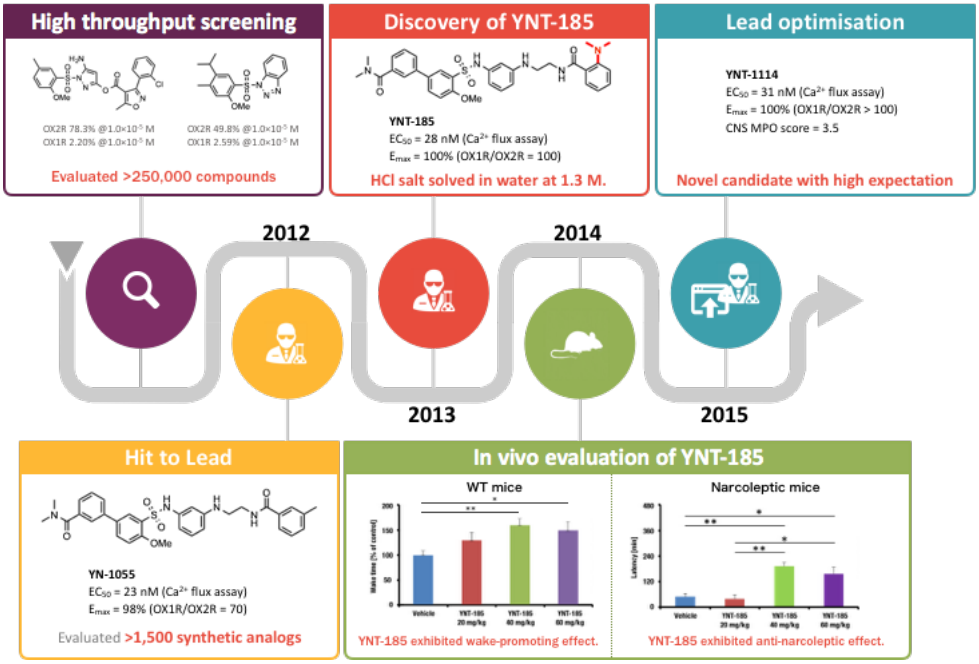


Figure 7: Development of OX2R selective agonists

[10] Identification of somnogenic natural compounds and elucidation of their mechanisms (Urade Lab)

(Papers underscoring the research achievement above and their brief account: [Appendix 2-1] 20-27)

We are focusing on the development of sleep promoting substances from foods or traditional medicines (Figure 8). We first screened hundreds of natural products (extract or major ingredient) by monitoring locomotor activity with passive infrared sensor. Compounds showed significant decreasing (or increasing) locomotor activities were measured sleep by monitoring electroencephalogram (EEG) and electromyogram (EMG) combined with locomotor activities. We analyzed the EEG and EMG data by the automated analytical system "Sleep sign" developed by collaboration with company. We identified sleep promoting 8 compounds and 1 extract (20-27).

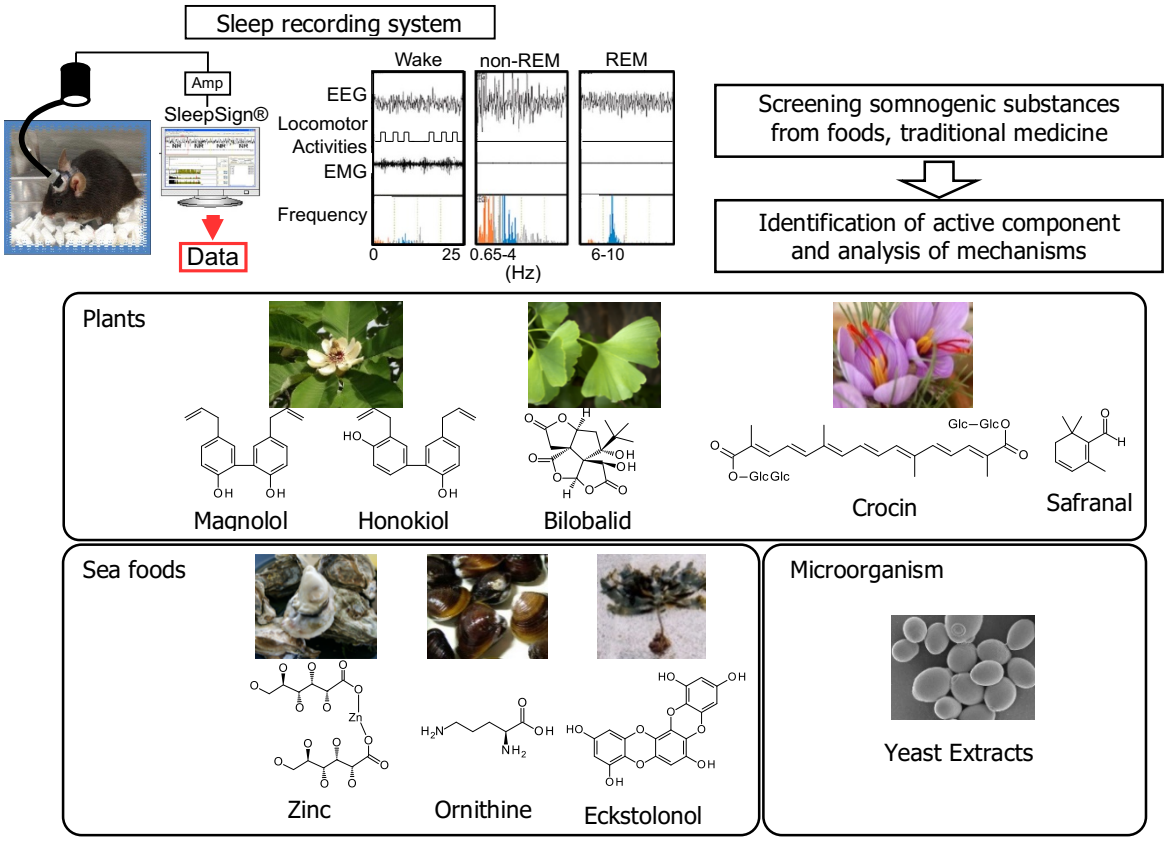


Figure 8: Screening for somnogenic natural compounds from foods and traditional medicine.

2-2. New Challenges

Describe the new challenges befitting a WPI center that have been undertaken.

As described in "**1. Summary of State of WPI Center Project Progress**," implementation of the translational research is our challenge to establish "sleep science" (or to create the new interdisciplinary domain). We aim at translating achievements in basic biology and pharmaceutical science to experimental medicine and/or clinical research. To enforce the translational research, our major efforts have been dedicated to increase and expand collaboration/research alliances with outside groups including groups in University of Tsukuba, the satellites, external research institutions, and even research groups in industries. We provide the following examples of the collaborative translational research programs as New Challenges.

(1) Study of the effect of the orexin antagonist on physical and cognitive functions (with Faculty of Health and Sport Science, University of Tsukuba)

The purpose of this collaboration is to investigate effects of Suvorexant, a novel orexin receptor antagonist, on physical and cognitive functions with a nocturnal forced-awakening condition in healthy men. A randomized, double-blind, placebo-controlled and crossover PSG study is planned to compare effects of the orexin antagonist with a $GABA_A$ agonist with similar PK profile, Brotizolam. At the time corresponding to C_{max}, subjects are forced to get up and subjected to choose reaction time task, postural sway, pegboard test and color-word matching Stroop task. The study is ready to be conducted and we are waiting for the approval from ethical committee.

(2) Exploring the somnogenic target of thalidomide (with Tokyo Medical University)

Thalidomide was marketed as a "miracle" sleep pill in late 1950's, but quickly withdrawn due to its severe teratogenicity, which unfortunately produced thousands of victims. The drug was clinically reintroduced as an anti-tumor agent in late 1990's. Cereblon, a component of the ubiquitin ligase pathway, was recently identified as the target for thalidomide's teratogenic and anti-tumor activities. However, mechanisms for the somnogenic action of thalidomide remain unknown. In order to examine

whether cereblon also mediates sleep-inducing effects of thalidomide, we generated gene knock-in mice possessing a thalidomide-resistant mutant allele of cereblon, and confirmed that cereblon-dependent ubiquitination was resistant to thalidomide in these mice. We then found that these mice are equally sensitive to thalidomide's somnogenic effects as compared with wild-type controls. Our results indicate that thalidomide's teratogenic and somnogenic effects could be dissociable, and point to a strategy for discovering new sleep inducing drugs.

(3) Screening of true short sleeper individuals and families for human genetic studies (Akita University Graduate School of Medicine)

Using a questionnaire, we recruited 10 candidates of short sleeper from 600 students in Akita University. All subjects are in good conditions without any medical problems and take no medicine influencing their sleep. We asked them to keep sleep diary for 8-19 days, and selected 7 subjects whose average full-sleep time is less than 5.5 hours. Further, we measured their sleep conditions for 8-14 days by using Actigraphy as well as sleep diary, and identified 6 subjects showing their average full-sleep times less than 5 hours. Interestingly two cases among them were found to have a family history of short sleeper phenotype. We now prepare for a study of human molecular genetics.

(4) Development of algorithms and software for fully-automated sleep/wakefulness stages analyses (with Center for Computational Science, University of Tsukuba)

We have developed algorithms and software, named exFASTER, to automatically analyze sleep/wakefulness stages of mice using EEG and EMG signals. We found several drawbacks of the software named FASTER, reported by Sunagawa *et al.* in 2013. We thus improved it in consideration of proximal stage shifts and experimentally confirmed that the modified software, exFASTER, showed higher accuracy in the stage analyses of mouse EEG and EMG datasets. We aim to develop software for fully-automated human sleep/wakefulness staging.

(5) Study of the effect of body-pressure dispersion of a mattress on sleep (with Nishikawa Sangyo Co.)

We study effects of body-pressure dispersion of a mattress on sleep. With healthy young male individuals, we conducted a random crossover study of mattresses showing different body-pressure dispersions by means of polysomnography. A mattress commonly used in the medical institution and the nursing homes were used as the control, and a mattress designed for higher body-pressure dispersion was used for the intervention. In the preliminary crossover study, the episode duration of slow wave sleep was found significantly longer with the intervention than that with the control. We are planning further to develop the study design.

2-3. Joint Research Advanced

Describe the joint research that the Center has undertaken with research organizations in and outside Japan.
・In Appendix 2-3, list and describe the cooperative research agreements that the Center has with other organizations.

(1) University of Texas Southwestern Medical Center (UTSW)

Yanagisawa had served as a professor/principal investigator for 20 years at UTSW, one of the best biomedical campuses in the U.S. We thus maintain a close relationship with it as one of IIIS satellites. He concurrently serves as Professor at the University of Tsukuba and UTSW as a joint appointment (95:5) between both universities and intellectual property rights of the Director will be succeeded by both universities based on this ratio, according to the collaborative research agreement for him. The four Satellite PIs (J. Takahashi, R. Greene, C. Green and Q. Liu) belong to UTSW and we have concluded collaborative research agreements or sponsored research agreements to provide them with research funds from the WPI subsidy. In the collaboration with them, we identified a novel signaling pathway of sleep/wake regulation by mass spectrometry and a novel molecular mechanism regulating innate fear by forward genetics, as described in 2-1.

(2) University of California, Berkeley

Since 2014, we have recruited Yang Dan as Satellite PI, engaging in joint research into the analysis of intracerebral neural circuits of sleep-awake control. We are planning tie-ups while developing analytical technology such as the optrode method, etc. to demonstrate power in analyzing nerve cell network.

(3) Akita University Graduate School of Medicine

We have concluded collaborative research agreement with Tetsuo Shimizu, a psychiatrist in Akita University Graduate School of Medicine and engaged in collaboration in the pathological study of sleep disorders such as narcolepsy, human genetics, etc. To facilitate human molecular genetic study of short

sleepers, six short-sleeper candidates were screened for and confirmed by analysis with ActiGraph as described in 2-2.

(4) RIKEN Brain Science Institute

We have been conducting joint research with Dr. Shigeyoshi Itohara of the RIKEN Brain Science Institute, Laboratory for Behavioral Genetics, on the study to elucidate the physiological significance of sleep toward developing therapy for mental disorders. In Dec 2015, one of the research results was jointly published in Science as described in 2-1.

(5) Ibaraki Prefecture/Ibaraki Prefectural Medical Center of Psychiatry

We conducted a joint project with the Hospital Management Division of Ibaraki Prefecture intended to promote clinical research into sleep disorders (sleep apnea syndrome in particular). A medical specialist of sleep apnea syndrome, M. Satoh was recruited as PI to conduct the clinical research at Ibaraki Prefectural Medical Center of Psychiatry to study involvement of breathing disorder during sleep in developmental disorder. Thanks to the donation of a venture company, Seven Dreamers Laboratories, Satoh Lab will be expanded to Clinical Sleep Medicine Lab in FY2016 to cover a broad scope of translational research. A successor to Dr. Satoh should be found for the clinical research at the Medical Center of Psychiatry.

(6) Graduate School of Pharmaceutical Sciences, Kyoto University

Since July 2015, we have installed Hitoshi Okamura, the Department of System Biology, Kyoto University Graduate School of Pharmaceutical Sciences as Satellite PI and concluded a collaborative research agreement. The objective of this collaboration is to fish out genes regulating jet-lag by ENU mutagenesis screening.

(7) Center for Genomic Medicine, Kyoto University

In December 2015, we concluded a collaborative research agreement with Dr. Fumihiko Matsuda, the Center for Genomic Medicine, Kyoto University to commence joint research to bridge our mouse forward genetics to a human genome epidemiological study, as a part of our translational research. In collaboration with us, they are also screening for individuals with extreme sleep phenotype by using actigraphy data from their large-scale Nagahama cohort study.

(8) JAXA Space Biomedical Research Office

The Grants-in-Aid for Scientific Research for Research into an Innovative Area: "Integratedly Understand the New Controlling System of Life from Space Point of View," which was jointly applied for by 11 groups including us, featuring Astronaut S. Furukawa as the area-representative, was adopted in FY2015. While we develop an automated sleep diagnosis system and evaluate insomnia treatment drugs in this project, we will also join the closed-environment stress test to be conducted in the isolation/confinement facility in JAXA, taking charge of sleep diagnosis.

In addition to the above, we are conducting five more joint research with universities or non-profit organizations under collaborative research agreements, and the details are listed in the Appendix 2-3.

2-4. Appraisal by Society and Scientific Organizations

Describe how society and/or scientific organizations in and outside Japan have recognized the Center's research achievements.
In Appendix 2-4, list the awards received and invitational lectures given by the Center's researchers.

(1) Appraisal in the US

Director Yanagisawa has been elected as a regular member of the National Academy of Sciences, and Joseph Takahashi (Satellite PI) has been also elected as the Institute of Medicine (IOM). Yanagisawa won the Walter B. Cannon Memorial Award: Physiology in Perspectives from the American Physiological Society in 2014. Joseph Takahashi and Yang Dan are Investigators of the Howard Hughes Medical Institute (HHMI) and have established themselves at the top of their research fields worldwide.

(2) Representative appraisals in Japan

Major awards are listed in Appendix 2-4. In short, Yanagisawa (2013) and Fukamizu (2015) honored with the Jokichi Takamine Award from the Society of Cardiovascular Endocrinology and Metabolism, Sakurai awarded the Prize for Science and Technology of the Commendation for Science and Technology by MEXT (research division) in 2013, Junichi Hayashi presented with the 24th Tsukuba Award in 2013 and Nagase successively honored with the Invention Prize of the National Invention Awards (2013) and Teiichi Yamazaki Award (2014). In the Japanese Society of Sleep Research, the academic society which leads hypnology in Japan, Yanagisawa was elected as the executive member in 2015.

In 2014, the world's first marketing approval was granted in Japan for an antagonist of orexin, a neuropeptide discovered in 1998 and found to be essential to maintain wakefulness by Yanagisawa, as the "first in class" drug for insomnia. Yanagisawa was partly engaged in developing this drug as the discoverer of orexin and gave a number of lectures after its release. An orexin antagonist is expected to function as the insomnia treatment drug without the adverse effects such as dependency, resistance, etc. of $GABA_A$ receptor agonists which have been used for many years as sleep-inducing drugs. The advantage received wide domestic and international media coverage and highly evaluated.

Apart from the above, as an appraisal from public, the fact we always get media attention as in TV programs such as TBS "Yume-no-Tobira＋," NHK "Science ZERO," etc. and in journals as "Nikkei Science," "Medical Asahi," "AERA," etc. is worthy of special mention. The prevalence of insomnia was 14% in the 2008 survey, rising as high as 30% in elderly population in particular and there is significant social concern over sleep problems. Attention from the media is considered to reflect it.

2-5. Center's Research Environment including Facilities and Equipment

Describe the Center's research environment including facilities and equipment and the state of its utilization.

The design of the new research building was completed in Nov 2013, construction started in Feb 2014, construction of the major part (6,000 m^2) supported by the MEXT subsidy completed in Mar 2015. Subsequently, construction of the additional part covered by in-house funding was completed, IIIS Building with 6 stories, total floor space of 8,000 m^2 (including 2,000 m^2 self-funded) was completed in June 2015. Relocation was completed in Aug 2015, and laboratories that were scattered around the university campus are now under the same roof, to form the leading global research center of sleep science covering the scope from molecular genetics, neuroscience, pharmaceutical science, clinical sleep physiology, and to experimental medicine. In Mar 2016, landscaping including the parking lots for 100 cars was also completed.

＜Floor plans＞

1F Common area	Entrance hall, auditorium, lounge, meeting rooms, Director's office, administration office, machine/electric room
2F Lab/office area	Mass spectrometry room, microscope room, cell culture room, low-temperature room, common equipment room, biochemistry/molecular biology labs, PI offices, lab office
3F Lab/office area	Clinical sleep physiology lab, cell culture room, sample preparation room, reagent preparation room, biochemistry/molecular biology labs, PI offices, lab office
4F Lab/office area	Chemical analysis room, NMR room, meeting room, chemistry labs, PI office, lab office, spare space for future expansion
5F Animal experiment area	Animal holding rooms, behavioral labs, sleep recording rooms, animal physiology lab, dissection room, special microscope room
6F Animal breeding area	Animal breeding rooms, cleaning and sterilization room, cage storeroom, material storage, waste storage, animal management office
RF	Solar power panel, air-conditioning equipment, exhaust equipment (scrubber, etc.)

Centering around an open ceiling space with a symbolic spiral staircase linking floors one to four, lab offices, lounges and auditorium are arranged in migratory fashion, so that researchers can naturally meet, interact and influence each other to further boost intelligent inspiration from various disciplines and fields of research and accelerate the fusion research. Five formative art works created by the joint project with artists in Faculty of Art and Design, University of Tsukuba are arranged/exhibited at each location in the building to stimulate the intellectual curiosity of researchers and symbolize the fusion (the costs of producing and exhibiting these art works were covered by donations.).

Concerning the design of experimental areas from the 2nd to 4th floors, researchers actively participated in it from the initial planning and their feedback was sufficiently taken into account. Functional and efficient experimental environments were successfully created by zoning according to the nature and purpose of experiments. For example, an experimental area on one floor is divided into 3 categories; the "lab" zone with benches exclusive for respective laboratories, the "lab support" zone where measuring equipment and/or analytical equipment are mostly arranged, and the "share support" zone where common equipment and functions are arranged.

The animal breeding/experimental areas, which require high levels of cleanliness and security, are located on the upper floors (5, 6F). These areas are comprised of clean rooms with state-of-the-art air conditioning system featuring airtight barriers established with room pressures controlled section by section, as well as individually configurable temperature and humidity. The breeding area on the sixth floor includes the cleaning and sterilization room with two large autoclaves and a rack washer, and breeding rooms with an automated water feeder, RO (reverse osmosis membrane) water-purifying apparatus and a device for producing weakly acidic sterilizing water, which can accommodate up to 6,000 IVC cages for mice/rats, capable of breeding tens of thousands of the same. The experimental area on the fifth floor also includes seven sleep recording rooms and six behavioral labs with the equipment for sleep/behavior analysis.

The major state-of-the-art equipment installed to date includes a large-scale electroencephalograph system, a fiber-optic confocal microscope for experimental animals, a high-resolution mass spectrometer (Orbitrap Fusion), 3D micro X-ray CTs for experimental animals (R_mCT2-SP), bioluminescent/ fluorescent *in vivo* imaging system (IVIS) for small animals, multiphoton excitation imaging systems (Zeiss Axio), FACS (BD 4LS), and slide scanners (NanoZoomer-XR).

The construction site (about 200,000 m^2) provided by the university for the new research building had been reserved as nature conservation greenery for over 40 years since University of Tsukuba was established. The landscaping was thus planned to minimize deforestation and to incorporate existing geographical features, a creek, a pound, slopes and woods into the design as much as possible. The spacious wood deck projecting toward the pound and the promenade running along the same around the research building are venues for researchers to relax and interact.

2-6. Non-WPI Project Funding

Describe the results in securing non- WPI project funding.

・In Appendix 2-6, draw of graph showing the Center's transition in securing non-WPI project funding and list external funding warranting special mention.

FY2012

Competitive research funds acquired by the core group amounted to 451,920,000 yen for the Funding Program for World-Leading Innovative R&D on Science and Technology (FIRST) project and 1,520,000 yen of other external funds, totaling 453,440,000 yen (portion allocated to FY2012).

FY2013

Competitive research funds acquired by the core group totaled 461,460,000 yen (portion allocated to FY2013), including 397,620,000 yen for the FIRST project and 63,840,000 yen of other external funds.

FY2014

Competitive research funds acquired by the core group totaled 177,930,000 yen (portion allocated to FY2014). Facing the end of the FIRST project, we actively applied for external funding programs and the total amount excluding FIRST tripled as compared to the previous year. In terms of Grants-in-Aid for Science Research, 12 programs including Yanagisawa's Scientific Research (S) were newly accepted, and 19 programs were supported to obtain 97,843,000 yen in total.

FY2015

Competitive research funds acquired by the core group totaled 282,070,000 yen (portion allocated to FY2015), approximately doubled in comparison with those in the previous year. A new project of Grants-in-Aid for Scientific Research for Research into an Innovative Area, for which we applied in cooperation with Space Biomedical Research Office, JAXA and 10 groups, was newly accepted, and the number of projects supported by Grants-in-Aid Scientific Research reached 28 including other new projects and continuous ones. For a further increase of funded projects in FY2016, we encouraged all qualified researchers to apply for Grants-in-Aid Scientific Research as in the previous year. We provided them with strong supports, such as having applications not only in Japanese but also in English checked by the faculty members in the administration department, and a total of 40 applications were submitted. PIs were ranked high on the amounts of external funding obtained by faculties in the university, and Center Director Yanagisawa, Urade and Hayashi were commended under the "reward system to give incentives for external funds allowing overhead costs" with recognition for financial contribution to the university.

From FY2015, we initiated research collaboration with a global pharmaceutical company. Further, we started two more collaboration programs with the local government and a bedding manufacturer; a special joint research project with Ibaraki Prefecture on treatment for sleep apnea syndrome

(75,000,000 yen/five years), and joint research program with Nishikawa Sangyo Co., Ltd. on "the effect of body-pressure dispersion of a mattress on sleep" in collaboration with Tokuyama, a collaborative PI of the Faculty of Health and Sport Sciences, University of Tsukuba (30,000,000 yen/two years). The research alliance programs have increasingly contributed to the research funding.

In addition, it has been agreed recently that from FY2016 we shall start collaboration with Sumitomo Dainippon Pharma Co., Ltd. to find a therapeutic agent using a model mouse of REM sleep behavior disorder (RBD) developed by Yanagisawa (21,600,000 yen/two years). Moreover, leveraging donations from Seven Dreamers Laboratories (36,000,000 yen/two years), we plan to expand Satoh Lab as an endowed research department (Clinical Sleep Medicine Lab).

2-7. Applications of research results

Describe the applications created from research results, their effect in spawning innovation, intellectual properties (IPs) obtained, and joint research activities conducted with corporations, etc.

Patent applications

We filed 10 patent applications as listed in the following table. The applications are all commensurate with the research strategy of IIIS, which involves elucidating the sleep-awake regulation mechanisms and the pathogenesis of sleep disorders and developing a new treatment method. Due to the nature of our research subjects, sleep science, the major means of practical application of research results include licensing of patents to pharmaceutical/diagnosis companies. At present, patent No. 10 in the table have been licensed to Toray Industries, Inc. and they are examining the feasibility of its medical application. We are engaging in negotiation with multiple companies for licensing of No. 3 and 8 below.

No.	Title of invention	Inventor	Date	Application number
1	Sulfonamide derivative or its pharma-ceutically acceptable acid addition salt	Nagase H, Nagahara T	Dec 12, 2013	Patent application 2013-257523 (PCT/JP2014/082961)
2	Preventive remedy for septicemia	Irukayama Y, Yanagisawa M, Ogawa Y	Mar 28, 2014	Patent application 2014-067451 (PCT/JP2015/ 59548)
3	Nalfurafine containing preparation for topical application	Nagase H, Saiki K, Shimoyama J, Tada M	Sep 30, 2014	Patent application 2014-201237 (PCT/JP2015/ 77741)
4	Orexin receptor antagonist	Nagase H, Irukayama Y, Ogawa Y, Miyamoto M, Nitta K	Oct 31, 2014	Patent application 2014-222969
5	Sulfonamide derivative or its pharmaceutically acceptable acid addition salt	Yanagisawa M, Nagase H, Irukayama Y, Saito T	Feb 19, 2015	Patent application 2015-031041 (PCT/JP2016/054700)
6	Morphinan derivative	Nagase H, Fujii H, Saito A, Nakata E, Hirose M, Oi I, Hayashida K	Mar 17, 2015	Patent application 2015- 54079 (PCT/JP2016/ 58475)
7	Sulfonamide derivative or its pharmaceutically acceptable acid addition salt	Nagase H, Yanagisawa M, Saito T, Kutsumura N, Irukayama Y	Jun 12, 2015	Patent application 2015-119785
8	Nalfurafine containing transdermal absorption cataplasm	Nagase H, Tada M, Yashima M, Saiki K	Jun 24, 2015	Patent application 2015-126282
9	Sleep state automatic judgment system and judging method in consideration of individual difference	Yanagisawa M, Satoh M	Oct 28, 2015	US provisional application (Application #: 62/247,329)
10	Morphinan derivative and its medical use	Nagase H, Yamamoto N, Irukayama Y, Saito T	Oct 29, 2015	Patent application 2015-212553

Joint research with companies

The joint research projects between IIIS researchers and the companies described below, also aim at practical applications of research results (seeds) obtained at IIIS to solve the problems of sleep disorders. In particular, we focus on translational research such as drug discovery in cooperation with pharmaceutical companies and clinical research on human sleep in collaboration with sleep-related companies/research institutions.

(1) Merck Sharp and Dohme (MSD)

From FY2015, we started the joint research project with MSD.

(2) Sumitomo Dainippon Pharma Co., Ltd.

We are starting exploratory research of treatment for REM sleep behavior disorder (RBD) using our

original RBD model mouse with Sumitomo Dainippon Pharma. We will screen and evaluate chemical compounds of Sumitomo Dainippon Pharma using this model, which has been validated with the orexin antagonist as described in 2-1.

(3) Fujifilm Corporation

From 2013 to 2015, within the framework of ”excellent sleep consortium” as a consignment study of the Ministry of Agriculture, Forestry and Fisheries, Y. Urade engaged in the joint research of ”pharmacokinetics of the sleep-improving ingredient and elucidation of a sleep-inducing mechanism” with Fujifilm and found the sleep-inducing action of zinc. Results of the collaboration with Fujifulm since his previous position in the Osaka Bioscience Institute (OBI) have been already implemented as a product marketed by them, the sleep-aiding supplement “Suttone.”

(4) Lion Corporation

We have conducted the ongoing joint research with Lion to “elucidate the mechanism of action of sleep quality-improving material.” Urade’s collaboration with Lion since his previous position in OBI resulted in Lion’s marketed product, the sleep-aiding supplement “Gussumin.” Urade has continued the collaboration with Lion to elucidate its mechanism of action.

(5) Nishikawa Sangyo Co., Ltd.

In FY2015 we started the joint research with Nishikawa on the “effect of body-pressure dispersion of a mattress on sleep.” Although experiences on a daily basis reaffirm the significant impact of bedding on sleep, given the lack of scientific research and validation, we aim to objectively evaluate the effect of body-pressure dispersion of a mattress on sleep. With mattresses showing various body-pressure dispersions, their effects on sleep are studied by using polysomnography. A preliminary result has suggested a potential parameter as a marker for sleep quality.

In addition to the above, we are conducting four joint research programs with for-profit organizations under collaborative research agreements, and the details are listed in the Appendix 2-3.

2-8. Achievements of Center’s outreach activities

・In Appendix 2-8, list and describe media coverage resulting from press releases and reporting.

Annual IIIS Symposia

Since inaugurating the institute in FY2012, we have hosted the international symposium every year (four times to the end of FY2015) and invited topnotch scientists from home and abroad in order to introduce the latest achievements in sleep research and relevant fields to researchers in Tokyo/Tsukuba community as well as the general public. Further, the symposium is aimed at promoting vigorous communication/interaction of IIIS members with outside (domestic and foreign) scientists in sleep science. For details, see the paragraph of “4-2-1. Organization of the international workshop.”

IIIS Seminar series

To introduce cutting-edge studies/methodology in sleep science and expand our research network, we host the IIIS Seminar Series, where we invite domestic and international researchers in sleep/neuroscience fields almost every other week; 76 seminars have been conducted since the inauguration in December 2012. This seminar series is understood not only as an opportunity to hold lectures but also as to assess faculty and postdoctoral fellow candidates. Moreover, since the lecturer is given the opportunity of individual interviews with all PIs, the exchange of research ideas and the expansion of human network are anticipated. The seminar is open and, in addition to IIIS members, many researchers/students in relevant fields in the university and other public/for-profit organizations participate in the seminar. The seminar series significantly boost the recognition of IIIS.

Super Science High School (SSH) Festival

We have opened the booth jointly with other WPI centers at the annual national meeting of SSH every year since FY2013, and explained about IIIS and its activities. This activity served as a trigger to close interaction/cooperation between IIIS and a few high schools. Especially, students in Udo High School, the SSH in Kumamoto are very much interested in sleep and decided to conduct a large-scale experiment, in which the whole school take a nap after lunch and see its effects on performance of students. It continued in FY2015 and could be described as one of the successful cases of our outreach activity.

WPI Joint Symposia

We have participated in the WPI Joint Symposia since FY2013 and developed our outreach activity, mainly targeting high school students, by setting up the booth and giving lectures. In FY2013, Center

Director Yanagisawa gave the keynote lecture, "Solving the mystery of sleep" at the WPI Science Talk Live, while in FY2014, Lazarus presented his research achievements at the booth.

AAAS Annual Meeting

In FY2013 and FY2014, we participated in the annual meeting of the American Association for the Advancement of Science (AAAS) and presented the research activities of IIIS to wide-ranges of participants, including scientists, policy makers, administrative officers, domestic and international media, the general public and other participants.

Lectures open to the public, Science Café, etc.

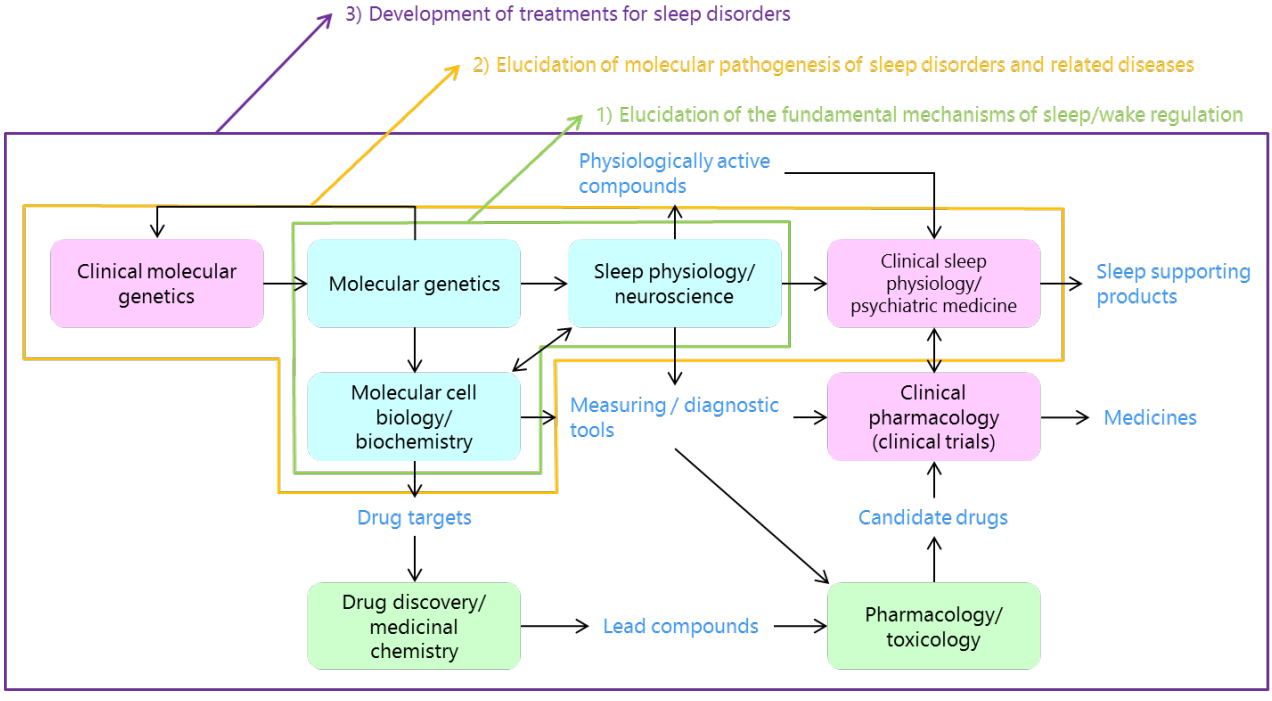
Yanagisawa and other IIIS PIs gave lectures at the International Congress Center Epochal Tsukuba and Ibaraki Prefectural Culture Center in FY2014, to the public to enlighten the importance of sleep science and explain the recent achievements. All lectures attracted people even in weekdays. Furthermore, we offered classes for children, high school students and Science Café on an irregular basis. The events for high-school students in particular are very popular and some high schools request regular visits.

3. Interdisciplinary Research Activities (within 2 pages)

3-1. State of Strategic (or "Top-down") Undertakings toward Creating New Interdisciplinary Domains

The research objectives of the IIIS are: 1) Elucidating the fundamental mechanisms of sleep/wake regulation, 2) Elucidating molecular pathogenesis of sleep disorders and related diseases and 3) Developing treatments for sleep disorders.

To achieve these objectives, it is required to integrate the three research fields, basic biology, experimental medicine and pharmaceutical science, as shown in the diagram below, to establish a new interdisciplinary domain "sleep science" which would be the comprehensive life science field focusing on sleep.



Accordingly, IIIS has been established as an independent research organization reporting directly to the University President, without belonging to any existing faculties such as Faculty of Medicine, Faculty of Health and Sport Sciences, Faculty of Life and Environmental Sciences, or Faculty of Pure and Applied Sciences. The positioning of IIIS in the university ensures performing the interdisciplinary research activities under the leadership of the Center Director. The Center Director is a physician with an intensive research background in molecular pharmacology. He discovered orexin and is also known as a pioneer of

neuroscience of sleep. His leadership is a crucial driving force to create "sleep science."

		Liu	Tak	TSa/ MSa	Hay	Laz	RGr/ Vog	CGr	Dan	Ura	Oka	Yan/ Fun	Nag	Sat/ Tok	Shi
Basic Biology	Molecular genetics	✓	✓								✓	✓			
	Biochemistry	✓	✓							✓	✓	✓			
	Molecular cell biology	✓	✓	✓	✓					✓	✓	✓			
	Sleep physiology		✓	✓	✓	✓	✓	✓	✓	✓	✓	✓			
	Neuroscience		✓	✓	✓	✓	✓	✓	✓	✓	✓	✓			
Pharma-ceutical Science	Pharmacology									✓	✓	✓	✓		
	Drug discovery											✓	✓		
	Medicinal chemistry												✓		
Clinical Medicine	Clinical sleep physiology													✓	✓
	Clinical pharmacology													✓	✓
	Psychiatric medicine														✓
	Clinical molecular genetics														✓

Liu: Qinghua Liu
Tak: Joseph S. Takahashi
TSa: Takeshi Sakurai
MSa: Masanori Sakaguchi
Hay: Yu Hayashi
Laz: Michael Lazarus
RGr: Robert W. Greene
Vog: Kaspar Vogt
CGr: Carla B. Green
Dan: Yang Dan
Ura: Yoshihiro Urade
Yan: Masashi Yanagisawa
Fun: Hiromasa Funato
Oka: Hitoshi Okamura
Nag: Hiroshi Nagase
Sat: Makoto Satoh
Tok: Kumpei Tokuyama
Shi: Tetsuo Shimizu

However, the leadership is not sufficient for fostering the interdisciplinary research. As seen in the table above, the teams in IIIS are comprised of PIs active in the field of basic biology as well as ones with sufficient experience and achievements in pharmaceutical science and experimental medicine, in order to ensure a team composition capable of interdisciplinary collaborations. Further, Kokubo with the experience of Senior Director of Drug Discovery Laboratories in a pharmaceutical company has been appointed to Administrative Director to coordinate translational research and the interdisciplinary collaboration.

To reinforce pharmaceutical science in the team, we have commenced joint research with a global pharmaceutical company. It is very difficult for academic institutions to conduct studies necessary for the pre-clinical development, *i.e.*, pharmacokinetics, pharamacodynamics and toxicology due to large resources specific for them. To fulfil the missing functions in pharmaceutical science and complete the framework of sleep science, the joint research with a pharmaceutical company is essential.

Moreover, to enhance research in the field of clinical sleep physiology as a part of experimental medicine, joint research with Faculty of Health and Sport Sciences, University of Tsukuba (Professor Tokuyama/Associate Professor Okura) and JAXA Space Biomedical Research Office (Astronaut Furukawa) has been undertaken as described in 2-2. Additional efforts of the translational research to establish the interdisciplinary domain includes the joint research with Kyoto University School of Medicine, Disease Genome Epidemiology (Professor Matsuda) to bridge our mouse forward genetics to a human genome epidemiological study, the collaboration with Center for Computational Science, University of Tsukuba to develop software of EEG analysis for fully-automated human sleep/wakefulness staging, and the joint development of portable EEG measuring devices with Center for Cybernics Research, University of Tsukuba.

A key element for success in creating the interdisciplinary domain, sleep science would be outcomes/performances of the collaborations among various groups. We thus nominate a project manager for each joint research project to secure good communication and smooth implementation of respective project. The University Research Administrator (URA) assigned to IIIS from the URA Support Office of the university headquarters has been assigned to the project manager, taking care of 6 projects.

3-2. State of "Bottom-up" Undertakings from the Center's researchers toward Creating New Interdisciplinary Domains

As mentioned above, creating a fusion research across different research fields is a difficult task to be performed by a single lab alone, and collaboration/joint research between respective labs even within

the core group in IIIS is essential. Until last August, however, the respective laboratories were dispersed over five locations on the campus of University of Tsukuba (TARA Center, Project Research Building, Health and Medical Science Innovation Laboratories, Building E of the University Hospital, Laboratory Animal Resource Center). Accordingly, communication among the members of respective laboratories was limited and there were very few opportunities for the bottom-up undertakings.

However, all the labs in the core group of IIIS moved to the new research building by last September and the labs conducting various studies across 3 research fields are literally operated under one roof. The offices of the respective labs are designed as open offices on the 2nd to 4th floors, facing the open ceiling space, in a structure facilitating interaction and communication in and between labs. The open offices on the 2nd to 4th floors are interconnected via a symbolic spiral staircase installed in the open ceiling space, to facilitate communication among members in labs located on different floors. Security points are only at the entrance from the entrance hall to the elevator hall and that from the auditorium to the lounge, and once past the security points, access between all laboratories is free and open.

In addition, to encourage interaction among the members of different labs, meetings such as 1. WIP meeting, 2. Journal club ”IIIS Dojo,” and 3. IIIS seminar are held frequently.

1. Work in Progress (WIP) meeting: After the relocation to the new research building, we started the WIP meeting, where research progress is reported by one researcher from the lab on duty from 9:00 every Wednesday. In principle, every PI and member of labs must attend, while the participation of external researchers is restricted. This allows the progress of studies in the respective labs to be shared in real time; fostering an environment to discuss bottom-up research collaboration and joint research.
2. Journal club (IIIS Dojo): Junior PIs have taken the initiative to establish the Journal Club, IIIS Dojo, where students and young researchers introduce papers related to sleep during the lunch break. As the name of "Dojo" suggests, the club is also intended as an opportunity of practicing English presentation as well as of studying current research for students and young researchers, a lively exchange of questions and answers is guaranteed. It is also an opportunity to share new approaches and methods of sleep research and one to seek research collaboration.
3. IIIS seminar: We conduct seminars, welcoming researchers who conduct interesting research basically from outside IIIS. This seminar is completely open, not only to teachers and students in the university but also researchers, etc. of companies outside the university, which makes it an opportunity to consult on the wider aspects of research collaboration. The lecturers of the seminar also have the chance to engage in individual interviews with all the PIs of the IIIS, before and after the seminar and the opportunity to consult on bottom-up research collaboration.

Among the PIs of IIIS, 9 PIs including Yanagisawa, the Center Director, Sakurai, Greene, Vogt, Satoh, Shimano, Matsuzaki, Shimizu and Sakaguchi are qualified in medicine. Conversely, a total of seven PIs and staff members have experience in implementing drug discovery research in a pharmaceutical company, including Nagase, Urade, Aritake, Kokubo, Fukusumi, Sasabe, Chikatsu and this personnel set-up also helps foster a climate where fusion research is created in three areas of basic biology, pharmaceutical science and experimental medicine in a bottom-up manner.

4. International Research Environment (within 4 pages)

4-1. International Circulation of the Best Brains

4-1-1. Results of International Joint Research (other than with the satellite)

Representative international joint research, which greatly contribute to enhance the global visibility of IIIS, are listed below.

(1) Northwestern University, U.S.

Yanagisawa/Funato Lab has been working with Dr. Ravi Allada (Edward C. Stuntz Distinguished Professor and Chair of Department of Neurobiology), to elucidate molecular mechanism regulating sleep/wakefulness and circadian behaviors, which is conserved between mammals and insects.

(2) Harvard Medical School, U.S.

Michael Lazarus has long been collaborating with Drs. Clifford B. Saper and Patrick M. Fuller in Beth Israel Deaconess Medical Center, Division of Sleep Medicine, to develop and use genetically engineered systems to define the cellular and synaptic basis by which the brain regulates sleep and wakeful consciousness, seeking to link the activity of defined sets of neurons with neurobehavioral and electro-encephalographic outcomes in behaving animals.

Kaspar Vogt works with Dr. Uwe Rudolph in the Program in Neuroscience, on GABAergic circuit specificity.

(3) University of Texas Southwestern Medical Center (UTSW)*, U.S.

Qinghua Liu is working with Dr. Bruce Beutler, the Nobel laureate in physiology or medicine in 2011, on a large recessive mouse screen to identify fearful and fearless mutant mice.
Liu is also working with Dr. Yonghao Yu in the Department of Biochemistry on proteomics and phosphoproteomics analysis of Sleepy and sleep-deprived mouse brains.
*Those collaborations are separate from the activities in UTSW as a satellite institute.

(4) University of Pittsburgh, U.S.

Hiroshi Nagase has been working with Dr. Sarah Ross in the Department of Neurobiology, on clarification of scratching behavior when feeling itch by characterization of B5-I neuron which releases dynorphin to attenuate the severe itch. B5-I neuron are innervated by menthol-, capsaicin- and mustard oil-responsive sensory neurons and are required for the inhibition of itch. These findings suggest that kappa opioid nalfurafine may be a broadly effective therapy for pathological itch (published an article on *Neuron* **82**, 573–586, 2014).

(5, 6) Massachusetts Institute of Technology (MIT), U.S.

Many other IIIS members are in a close collaboration with Dr. Matt Wilson in The Picower Institute for Learning and Memory, visiting his laboratory for academic exchanges as well as inviting him as a plenary speaker for the Annual IIIS Symposium.
Masanori Sakaguchi works with Dr. Edward S. Boyden, MIT Media Lab, on developing optogenetics tools and methods (Sakaguchi *et al*., *PLoS One* **10**(6): e0130163, 2015), which created another international collaboration with Dr. Brian Wiltgen, **University of California Davis Center for Neuroscience**, for optogenetic control for memory.

(7) Universite de Montreal, Canada

Nagase has been working with; Dr. Graceila Pineyro, Department de Psychiatrie on G protein activation, beta-arrestin recruitment of KNT-127 (delta opioid agonist which isolates the catalepsy and seizure different from another delta agonists). They are trying to clarify the reason for only KNT-127 has no catalepsy studying the contribution of beta-arrestin to the catalepsy and seizure.

(8) University of Toronto, Canada

Sakaguchi has been working with Dr. Paul W. Frankland in Neurobiology Laboratory at the Hospital for Sick Children, on the functional integration of adult-born neurons (Arruda-Carvalho *et al.*, *J. Neurosci.*, **34**(47):15793-803, 2014).

(9) Fudan University, China

Liu has been working with Dr. Jinbiao Ma, Department of Biochemistry on structural and functional studies of the RISC loading complex.

Lazarus has been collaborating with Drs. Zhi-Li Huang and Wei-Min Qu on the development and use of genetically engineered systems to define the neurobiological basis of sleep and wake, and have made key contributions to our understanding of the role of basal ganglia in sleep/wake regulation.

(10, 11) Korea Advanced Institute of Science and Technology (KAIST), Korea

Yu Hayashi is coordinating a collaboration on the roles of neuropeptides in cognition together with Dr. Seung-Hee Lee (Assistant Professor, Department of Biological Sciences). This collaboration also includes Dr. Chengyu Li, Principal Investigator, Institute of Neuroscience, Key Laboratory of Primate Neurobiology, CAS Center for Excellence in Brain Science, **Shanghai Institutes for Biological Sciences, Chinese Academy of Sciences, China.**

Sakaguchi is also in ongoing collaboration with Dr. Jin-Hee Han of KAIST.

(12) Institut national de la santé et de la recherche médicale (Inserm), France

Lazarus is working with Dr. Pierre-Herve Luppi to generate genetically engineered systems for the French lab to investigate brain circuits for the control of REM sleep.
Kaspar Vogt has been working with Dr. Marco Canepari on a novel imaging technology for submillisecond membrane potential changes from individual regions of single axons, dendrites and spines (*Adv Exp Med Biol*. **859**:57-101, 2015; *Front Cell Neurosci.* **8**:311, 2014).

(13) University of Zurich, Switzerland

Vogt has been working with Dr. Anne Marowsky in the Department of Pharmacology and Toxicology,

on inhibitory circuits in the amygdala. This collaboration resulted in the publication on *Front. Neural Circuits* **8**:27, 2014.

(14) The University of Edinburgh, England

Sakaguchi worked with Dr. Szu-Han Wang in the Center for Clinical Brain Sciences, on the mechanism of PTSD (Fujinaka *et al.*, *Mol. Brain* **9**:2, 2016), which suggested the importance of immediate care of the patients. This work is also a collaboration with Dr. Nohjin Kee in the University of Toronto, Canada.

(15) University of Melbourne, Australia

Nagase has been working with Dr. Tony Verberne in Clinical Pharmacology & Therapeutics Unit, Department of Medicine on 1) the effect of a potent non-peptidic OX2R agonist (YNT-185) on blood pressure, heart rate, adrenal sympathetic nerve activity and lumbar sympathetic nerve activity, 2) the effect of YNT-185 on glucose infusion rates in a rat hyperinsulinemic-euglycaemic model with glucose clamp.

4-1-2. State of Top World-level Researchers residing at the Center

Describe the participation of overseas Principle Investigators, the short-term stays of joint researchers, and the state of participation in symposiums sponsored by the Center.

・In Appendix 4-2, enter the number of researchers from abroad within the total number of the Center's researchers, and their annual transition

The overseas PIs actively participate in the research activities at IIIS. While Liu and Greene contribute to IIIS through their research activities in UTSW as Satellite PIs, they have also established their own labs as PIs in the core group of IIIS since FY2013 and engaged in lively research activity. Liu stayed at IIIS for 58 days during 6 visits to Japan in FY2013, 98 days over 6 visits in FY2014, and 108 days over 7 visits in FY2015. On the other hand, Greene stayed at IIIS for 20 days in 2 visits to Japan in FY2013, 17 days in 2 visits in FY2014, and 30 days in 3 visits in FY2015. They actively contribute to the management of IIIS by participating in the PI meeting held monthly, even when absent from the institute, via Skype from UTSW. They also actively participate in important events including the symposium hosted by IIIS and the annual site visit.

Other Satellite PIs mostly came to Japan at least once a year, stayed at IIIS for four to five days to hold meetings concerning the joint research and to attend the symposiums hosted by IIIS and the annual site visit.

For the trend of the ratio of foreign researchers (PI) as a proportion to all researchers (PI), refer to Appendix 4-2.

4-1-3. Utilization and Employment Situation of Young Researchers

Describe the utilization and employment situation of young researchers including postdoctoral researchers.

・In Appendix 4-3, enter the state of international recruitment for postdoctoral researchers, applications received, and selections made

・In Appendix 4-4, enter the percentage of postdoctoral researchers from abroad

・In Appendix 4-5, enter the state of postdoctoral researchers' employment

To recruit female PIs and young researchers including postdoctoral fellows, IIIS engages in international open recruitment by placing job advertisements on websites and journals such as the homepage of IIIS, Science, Naturejobs, Federation of European Neuroscience, Sleep Research Society-job board, Society for Neuroscience-Neurojobs, jREC-IN, American Society for Neurochemistry, etc. The number of applicants in each year is as shown in Appendix 4-3. As a matter of fact, since the most applicants did not meet the requirements imposed by IIIS, applicants through the international recruitment were not hired except for one Canadian researcher hired in FY2014. However, via the international network of PIs and Satellite PIs, we successfully recruited a foreign (Indian) researcher in FY2012 and 4 foreign researchers (1 French, 1 German, 2 Canadians) in FY2013. Additionally, in FY2014 we hired 3 foreign researchers (1 Russian, 2 Indians), and also in FY2015 we hired 3 foreign researchers (2 Chinese, 1 British) including a postdoctoral researcher from our satellite in UTSW. Apart from that, we also actively conducted recruitment at the opportunities such as the symposium hosted by IIIS, international conferences of the respective academic societies, etc. We also take advantage of the IIIS seminar series regularly inviting lecturers from outside as an opportunity to look for PI candidates among the lecturers (especially, young PIs and female young PIs).

4-1-4. Other

Describe the Center's policy for sending Japanese researchers overseas to gain international experience, and give examples of how the Center is working to create career paths for its researchers within a global environment of researcher mobility.

Looking at FY2015 overseas business trip track record of IIIS, of the 21 overall cases, 6 or third part of the total, were taken by young researchers. We are recommending more young researchers in the next fiscal year to actively take part in overseas exchanges.

To obtain budget to dispatch more young researchers overseas, we applied to "The Strategic Young Researcher Overseas Visit Program for Accelerating Brain Circulation" for FY2015, but failed to get the grant. We will thus try it again in FY2016.

4-2. Creating the Center's Environment

4-2-1. Holding International Research Meetings

Describe the results obtained from holding the Center's main international research meetings.

・In Appendix 4-6, enter the number of international research conferences or symposiums held and give up to two examples of the most representative ones.

IIIS Symposia (four times)

On March 27, 2013 at the International Congress Center Epochal Tsukuba, we hosted the first kick-off IIIS Symposium. All PIs and Junior PIs appointed at the inauguration as well as 2 keynote speakers gave lectures, and approximately 190 people including relevant researchers and students participated in the symposium. The symposium offered an opportunity for all members of IIIS and its satellites to get together and to confirm objectives and strategies of the joint research.

The second IIIS symposium was held on January 20, 2014 at the International Congress Center Epochal Tsukuba. As well as 6 invited speakers from Japan and abroad, PIs also gave talks, and approximately 150 people participated. After the talk at the 2nd IIIS symposium, we decided to welcome Yang Dan, University of California, Berkeley as a satellite PI.

We hosted the third symposium (September 24, 2014) as a joint symposium on "Homeodynamics in Clocks, Sleep and Metabolism" with Dr. Hiroki Ueda (The University of Tokyo Graduate School of Medicine/Laboratory for System Biology, RIKEN) and Dr. Joseph Bass (Northwestern University), inviting 24 domestic and international speakers. Over 230 participants attend the joint symposium held as Tokyo Translational Therapeutics Meeting at Ito Hall, the University of Tokyo, proving a great success.

The fourth IIIS symposium was held on February 26, 2016 at the IIIS Building for the first time since the new building has been launched. At this symposium, 11 invited speakers including many rising stars in sleep research presented their recent achievements, and the event attracted approximately 170 participants.

The 68th Fujihara Seminar

Supported by the Fujihara Foundation of Science, the 3rd IIIS symposium described above was followed by the 68th Fujihara Seminar at IBM Amagi Homestead, Ito, Shizuoka on September 25-27, 2014. As well as the lecturers invited to the symposium, many senior and young scientists in IIIS, the University of Tokyo and RIKEN in the relevant research areas participated in this three-day intensive workshop and engaged in a lively discussion.

Tsukuba Global Science Week (TGSW)

At Tsukuba Global Science Week, which is a campus-wide international exchange event of University of Tsukuba, two PIs gave lectures in FY2013. In FY2014, we held a session at TGSW where four high-profile researchers invited from the U.S. and IIIS PIs gave lectures on their recent achievements.

4-2-2. Support System for Overseas Researchers

University of Tsukuba has a department, "University of Tsukuba, Global Commons, International Exchange Support Office" (former Kasuga Plaza International Exchange Corner) which engages in livelihood support for foreign researchers and their families. They provide information on accommodation for foreigners in and outside the university and daily life in Tsukuba, and offer services including Japanese classes, proxy application of the certificate of eligibility (visa), some aid for various procedures and paperwork preparation, etc. Foreign IIIS researchers benefit significantly from the Support Office.

Japan International Science and Technology Exchange Center (JISTEC) operate the Tsukuba Office and IIIS concluded the agreement on support for foreign researchers with JISTEC. They offer the paid

service at minimal costs of attending to foreign researchers for the residence registration at City Hall, opening a bank account, etc. Further, many IIIS foreign researchers use the accommodation (Ninomiya House and Takezono House) managed by Projects to Support Living and Housing for Foreign Researchers/Management Services for International Accommodation of JISTEC.

Some researchers use the accommodation for foreign staffs on the campus of University of Tsukuba, in a highly convenient location and with well-organized support. The university plan to establish an international house and short-stay dormitory to accept students and short-term trainees from overseas as a private finance initiative (PFI) project, and it is expected that we will be able to improve the livelihood support for foreign researchers.

Conversely, in IIIS, we translate various forms into English, including documents of experimental plans in various regulatory applications and formats/documents related to employment, personnel affairs and general affairs. Other documents are also converted into English as necessary. We also support our foreign researchers by making sure all notices and announcements received from the university administration are translated into English from the original Japanese by the IIIS administration. In this way, our researchers are able to stay aware of university-wide as well as institutional information.

Following the relocation to the new research building in FY2015, we have equally assigned secretaries proficient in English to all labs to provide foreign PIs, researchers and students with sufficient supports.

5. Implementing Organizational Reforms (within 3 pages)

5-1. Operation carried out under the Center Director's Leadership

Describe the division of roles and authority between the Center and its host institution, and the state of the Center director's presence at the Center.

Decision-making

For important matters concerning the operation of IIIS, all decision-making was done in accordance to the Center Director's top-down approach. So that the intention of the Center Director takes effect quickly, organizational bylaws and other related regulations continue to be revised or enacted. By positioning IIIS as an independent research center in the university, wide-ranging autonomous operations, including personnel affairs, environmental improvement and expenditure are secured.

Concerning the authority given to the host institution, i.e., the appointment or dismissal of the Center Director, the President, University of Tsukuba introduced a combined-wage system that enables the cross-appointment to the university in March 2014. Based on it, the Center Director was officially employed by University of Tsukuba through the joint appointment with UTSW as of April 1, 2014. The effort ratio of the Director between University of Tsukuba and UTSW is 95:5 and his portion of intellectual property rights created by employee inventions is succeeded by both universities according to this ratio.

Principal Investigators' meeting (PI meeting)

Spearheaded by the administrative department, PI meetings were established to provide a periodic opportunity for PIs to openly discuss their opinions and concerns with the Center Director and to form consensus important matters concerning IIIS. In this meeting, the Director acts as the chairman and participants include all PIs and relevant staffs in the administration. The Administrative Director leads the administrative department and, based on the Director's policy, prepares a draft personnel plan, a draft budget plan, etc., to assist the Director. PI meetings are held once a month with video conferencing capability to allow the Satellite PIs running their labs in the core group (Liu and Greene) to also attend. The functions of the existing steering committee (deliberation on institutional organization/management, research plans, etc.) within IIIS have also been attached to the meetings. Junior PIs are also allowed to participate in the meetings, which gives a degree of motivation to the talented young researchers by granting them a forum for the management of their labs as independent researchers.

IIIS personnel committee

We established a personnel committee in IIIS to develop a system to appoint faculty members. The appointment system is different from the previous personnel system with less steps of the examination accelerated through intensive deliberations, i.e., 2 steps of the examination in total: the personnel committee and Administration Center Appointment Committee in the headquarters. Our system let us determine and appoint IIIS faculties under the leadership of the Center Director. The committee was

held twice in FY2015 (seven times in total), and the appointment of 1assistant professor and 1 visiting associate professor, and the promotion of 1 assistant professor to the associate professor were decided.

Physical presence of the Center Director in the University of Tsukuba

The records of the Center Director's attendance at IIIS were 143 days (58%) in FY2012, 211 days (86%) in FY2013, 226 days (93%) in FY2014, and 217 days (89%) in FY2015.

5-2. Administrative Personnel who facilitate the use of English in the Work Process

Composition of the administrative department

The administration is under the leadership of the Administrative Director, who has research management and strategy expertise accumulated during his assignment to Senior Director of the research institute of a pharmaceutical company. Supporting the Administrative Director is the Vice Administrative Director, who has served many years as a section head within the university headquarters. In this capacity, he is intimately familiar with the university administration operational procedures and campus affairs. The four teams within the administration are listed below.

General Affairs Team (3 members)

This team carried out work related to general affairs, human resources, hiring, business trips, office time management, etc. Two full-time university personnel having a long and deep experience with general affairs at the university (including the Vice Administrative Director, holding this concurrent post as Team Leader) were assigned from the headquarters to IIIS. Along with transferring a staff member with English proficiency from Research Strategy & Management team to provide support for foreign researchers joining IIIS, we also took advantage of Tsukuba's location as an international science city to contract Japan International Science and Technology Exchange Center (JISTEC) for additional support.

Accounting Team (3 members)

This team carried out works relating to budget management and enforcement, supply purchases, transfer of funds and goods domestically and internationally, etc. One full-time university personnel having a long history of familiarity with university budgeting and accounting serves as Team Leader.

Research Strategy & Management Team (3 members)

This team is charged with a wide range of work relating to budget planning, personnel planning, supports for competitive research funding application, project management, conclusion of contracts/agreements, support for patent application, report preparation, etc. A Ph.D. with experiences of drug discovery and expertise in contracts/negotiation built in the liaison office in research division of a pharmaceutical company serves as Team Leader. An URA assigned from the university headquarters has joined the team since June 2015 to reinforce its capabilities.

Alliance & Communication Team (2 members)

This team carried out works relating to public relations (news coverage, press releases and outreach activities), campus seminars, PI meetings, IIIS symposium planning and management, report preparation, etc. A Ph.D. holder with drug discovery experience in a pharmaceutical company as well as experience overseas serves as Team Leader.

Use of English as official language and employment of bilingual staff members

English is used as an official language at IIIS. A bilingual environment has been implemented, with 60% of administrative staff members fluent in spoken and written English (except for the people who have specific skills and/or experience that cannot be replaced by any other people). English is used as the official language for PI, WIP and other official meetings. In order to communicate frequently with overseas Satellite PIs, video conferencing/Skype systems have been equipped in 2 meeting rooms. Documentation is in English or bilingual as much as possible, except where it has to be in Japanese for external reasons.

IIIS hired people with overseas experiences and/or with an excellent command in English. English proficiency, especially the speaking abilities are judged at the job interview. This year a new staff member who is proficient in English was hired to assign her to a secretary for PIs so that we provide all PIs including foreign ones with even administrative services. Many administrative staff members regularly attend the PI meetings and vigorously participate in the discussions using English. Some staff members also join WIP meetings to receive updates on scientific knowledge and share the information on progress in studies in IIIS.

5-3. System Reforms and Their Ripple Effect within the Host Institution

Describe the following:
- Reforms to the Center's research operation advanced by way of the WPI Program's research-results evaluation system
- Reforms to the Center's operation made by introducing a merit-based salary system
- Ripple effects of the Center's system reforms within the host institution

Introduction of a system to evaluate research results and ability-linked salary system

This year, we promoted one of Junior PIs, who made remarkable achievements, from Assistant Professor to Associate Professor after the designated procedures (PI meeting, IIIS personnel committee, and Administration Center Appointment Committee in the headquarters) and approved a pay rise.
We have selected candidates of the advisory board members, as a part of our efforts to establish a system to evaluate research performance of PIs and link the appraisal to their salary. However, due to budget constraints (especially unexpected reduction of the WPI subsidy from the initial plan), we were not able to make selections of members and hold board member meeting. Conversely, from FY2015 at University of Tsukuba, a quantitative evaluation index concerning research performance in terms of published papers and writings, granted external funding, and research alliance with public and for-profit organizations, is being established. We are thus considering the index as an evaluation tool to build a system of merit-based compensation.

The salaries for the administrative staff members should be determined by the Center Director, based on the opinion of the Administrative Director. However, raises have been passed upon due to the budget constraints except for a few exceptions to remedy imbalances. The salary of a researcher invited from outside is decided by the Center Director, considering the duty at IIIS, research achievements and the salary at the previous job.

Authority over personnel matters and simplification of the appointment system

In IIIS, as described in 5-1, the personnel committee has been established and distinctive authority over personnel matters is assigned. Only 3 research centers in University of Tsukuba, i.e., IIIS, TARA Center and the Center for Computational Sciences, are given such authority over personnel matters. In particular, the appointment system of the IIIS is simplified to be comprised with two steps, namely the intensive deliberation at the personnel committee and the approval at Administration Center Appointment Committee in the headquarters, allowing speedy judgment and appointment by the leadership of the Center Director. University of Tsukuba now plan reorganization/restructuring of all the research centers according to the third mid-term plan. In future, our personnel system would serve as a good model for those of reorganized research centers in the university.

Joint appointment system

With the purpose of enabling the Center Director to occupy concurrent posts at University of Tsukuba and UTSW, the joint appointment system was newly introduced to University of Tsukuba in March 2014. At the same time, a Collaborative Research Agreement was concluded between University of Tsukuba and UTSW to determine terms and conditions of the research alliances spontaneously accompanied with the joint appointment. In response to the execution of the agreement, University of Tsukuba made the tenure appointment of Yanagisawa as of April 1, 2014. Subsequently, Liu was also employed from FY2014 under the joint appointment system, demonstrating, in University of Tsukuba, IIIS take the initiative in implementing the research alliance with overseas universities and offer model cases of the cross-appointment. Similarly, Vice Center Director, Sakurai, benefits from the combined-wage system to utilize multiple resources for the employment offered by the Faculty of Medicine, the Headquarters and IIIS, underlining the achievement of this system.

Tsukuba Short-term Study Program (TSSP)

To boost the international profile of the IIIS in sleep science, we are currently considering holding a workshop and offering short-term stay for training (internship) for young researchers. We conducted a questionnaire at the 3rd IIIS symposium in FY2014 to gauge interest and demand for a proposed training. Surprisingly 80% of the foreign researchers and students who responded to the questionnaire stated an interest in such training.

As for the short-term stay for training we have already accepted many trainees as described in Appendix 5-1. The system we have used to accept the trainees is the workshop type of Tsukuba Short-term Study Program (TSSP) of University of Tsukuba, which allows even short-term trainees to use the student dormitory at a nominal fee and requires no entrance and tuition fees. However, the term of the program was limited to less than 3 months. We were requested to accept undergraduate students as interns for the periods longer than 3 months by a few professors in China and Vietnam, but we could

not accommodate their requests. We thus consulted the Vice President in charge of student affairs for a solution. The student affairs division, University of Tsukuba reviewed the system and regulations, whereupon the bylaw for TSSP was revised in March 2016 and the stay for training within a year became possible. We plan to accept some students as interns under this revised program in FY2016, and this system is also expected to be utilized in other departments in future.

Operation of ARC Satellite

From October 2015, we started operating the animal facility (ARC Satellite) on the fifth and sixth floors of the new research building. We appointed Hiroyoshi Iseki, who is the assistant professor in S. Takahashi's Lab, to the Laboratory Animal Administrator managing ARC Satellite, as a part of the collaboration with our collaborative PI, S. Takahashi. We newly organized the ARC Satellite steering committee (chairman: the Center Director; members: the Administrative Director, the Laboratory Animal Administrator and relevant PIs), which decides on important matters, including the operation policy and rules, budgeting, investment and the charging system, by mutual consent. ARC Satellite is positioned literally as the satellite of the Laboratory Animal Resource Center (ARC) of the Faculty of Medicine, University of Tsukuba, and operated in collaboration with ARC. However, advanced animal breeding equipment such as individual ventilation cage (IVC) systems, zoning by cleanliness, acidic water for sterilization as an alternative to ethanol, have been introduced, and ARC Satellite serves as a pilot of new animal breeding techniques and facility in University of Tsukuba. As of March 31, 2,000 IVCs for mouse breeding are operating and a large number of genetically-modified mice (160 strains) are maintained.

Establishment of IIIS-TLO

As one of potential measures to ensure the continuous operation of IIIS after the completion of the WPI program, we are considering a system whereby license revenues of the intellectual property rights created by IIIS could be directly used to offset partially the costs of operating the Center. One of options of the specific method for implementing this idea is to establish a company to manage the IP rights and promote licensing them (tentative name: IIIS-TLO). We now discuss feasibility of the option with the university administration.

5-4. Support by Host Institution

Besides the state and effectiveness of the host institution's support for the Center, describe the Center's positioning within the host institution's mid- to long-term plans.

・In Appendix 5-1, describe specific support measures being taken by the host institution.

Positioning of IIIS in the mid-term plan of University of Tsukuba

During the third mid-term plan of University of Tsukuba starting from FY2016, the university aims to develop a globally unrivaled frontier research of both objectives, i.e., research to deeply seek truth and research for application contributing to society, in wide-ranging disciplines and research fields. To realize this objective, the university will make a plan of reorganization/restructuring/merger of all research centers and implement it during the period of the 3rd mid-term plan. IIIS is positioned as a pioneering model of the forefront research organization the mid-term plan targets.

The Research Promotion Department is fully aware of the need for Next-Generation Sequencing Core requested by IIIS, and is reviewing the plan we proposed, which features i) high-speed genetic analyses with next-generation sequencers and a high-throughput genotyping system and ii) a large capability of bioinformatics, to consider budgetary request for next fiscal year to MEXT.

Costs of constructing the new research building

The IIIS Building with six stories above ground and total floor area of 8,000 m^2 (including 2,000 m^2 self-funded) was completed in June 2015. It costed approximately 3.8 billion yen in total, including the costs of equipment, landscaping, etc. The expenses beyond those covered by the facility development subsidies of 2 billion yen kindly provided by MEXT were supported by in-house funding.

Tenure positions of PIs

In the third mid-term plan of University of Tsukuba starting from FY2016, it will set out a strategic framework of research resources over the entire university and plans to reallocate it, based on evaluation of research activities/achievements. Tenure positions will also be subjected to the reallocation. The President, University of Tsukuba, Dr. Nagata has committed himself to offer a tenure position to the PI that produces sufficient research achievements in IIIS by using the planned reallocation system, so that IIIS will survive as a World premier international research center beyond the end of the WPI program implementation period. Details shall be decided upon consultation between the Vice Presidents in charge of Research and Personnel Affairs.

Other resources provided by University of Tsukuba as support for IIIS

University of Tsukuba have provided IIIS with various resources as support. The provided support is equal to or greater than the support planned in the application for the WPI program, as described in the Appendix 5-1.

5-5. Others

5-5-1. Efforts to Foster Young Researchers (e.g., state-up funding)

Start-up funding in FY2012

To improve the research environment and equipment of their labs, we allocated start-up funding for PIs and Junior PIs invited from outside University of Tsukuba (Total amount: 31 million yen).

Start-up funding in FY2013

To launce their research launch programs, we provided PIs and Junior PIs invited from outside of University of Tsukuba with start-up funding. The plan of the funds was drafted by the Administrative Director and implemented based on the budget plan decided by the Center Director (Total amount: 63 million yen).

Start-up funding in FY2014

As start-up funding, especially for researchers who failed to acquire competitive funds such as Grants-in-aid for Scientific Research, etc., an internal grant system was introduced. Applications with a research plan were collected within IIIS and screened by faculty staff in the administrative department to secure fair and unbiased review. The research funds were provided based on a given priority (Total amount: 52 million yen).

Start-up funding in FY2015

To solve issues faced by foreign researchers in particular, i.e., difficulty in acquiring competitive funds such as Grants-in-aid for Scientific Research, we continued the internal grant system and invited applications with a research plan within IIIS. Like the previous year, to secure neutrality of screening, the screening was performed by the faculty staff in the administrative department. The funds were granted according to a given priority (Total amount 9.45 million yen).

Other efforts

After moving to the new research building in July 2015, we started the institution-wide weekly meetings all the IIIS researchers getting together to have progress reports (WIP meeting) and Journal Club (IIIS Dojo), in order to give young researchers and students opportunities of giving presentation and discussing beyond lab boundaries.

Many PIs and other faculty members in IIIS have been qualified for teaching and mentoring graduate students by respective graduate school in University of Tsukuba: ten from Graduate School of Comprehensive Human Sciences Majors of Medical Sciences, nine from the Graduate School of Comprehensive Human Sciences Master's Program in Medical Sciences, nine from the Ph.D. Program in Human Biology (HBP), and one from the Doctoral Program in Chemistry, Graduate School of Pure and Applied Sciences. In particular, it would be worthy of special mention that the unofficial lectures given by associate professor/junior PI, Vogt to meet the request of students in HBP, was adopted as official lectures in the curriculum of HBP (Neurobiology) from FY2016. Teaching and mentoring are good opportunity for Junior PIs and other young faculty members not only to contribute to education and training of a student but also to recruit her or him to their Labs and gain experiences of education and coaching.

5-5-2. Appointment of Female Researchers

・In Appendix 5-2, give the transition in the number of female researchers.

The number of women among researchers in IIIS is steadily increasing, and in FY2015, of the 32 researchers excluding PIs, 13 (41%) were women. Concerning PIs, in FY2014, Professor Yang Dang arrived at her post of Satellite PI and in FY2015, among 22 PIs, 2 (9%) were women. To ensure women-friendly work environment, although IIIS researchers are basically contract employees on single-year agreements, when PIs agree to their employment the following year, as an exception, they are allowed to take childcare leave and use short-time working for childcare to strive for operations according to their life stage.

Regarding the hiring of female PI into the core group, we have actively been recruiting with female

only PI advertisements on a number of websites and journals. The applications from female scientists are increasing, and we have conducted job seminars inviting 3 candidates to the IIIS seminar. At present, there have been no successful candidates; however, we will continue our recruitment efforts into the future. We also consider promotion to Junior PI from within IIIS to continuously strive to secure human resources while maintain the qualification.

6. Future Vistas (within 2 pages)

6-1. Future Policies and Plans for Advancing the Center's Operation and Project

Urgent agendas to realize the concepts of IIIS include the following:

1. Improving and maintaining facilities and equipment to achieve a "World Premier International" standard.
2. Further developing strength of IIIS such as capabilities of neuroscience in basic biological.
3. Filling gaps in research capabilities in pharmacokinetics, toxicology, etc., in pharmaceutical science.
4. Strengthening capabilities in clinical sleep physiology, human molecular genetics, etc., in experimental medicine.

In FY2015, we took measures against each agenda with the following solutions and major improvements were achieved toward realizing our concept:

1) A new research building was completed with considerable support from MEXT and University of Tsukuba and all Labs in the core group of IIIS were translocated to it, enabling cross-sectional research among the three research fields.
2) It was decided that Sakurai, Kanazawa University, who significantly contributed to the discovery of orexin and its functional analysis, should be transferred to IIIS as Vice Center Director as of April 1, 2016.
3) We started the collaborative research with a global pharmaceutical company, in which they agreed to share responsibility of pharmacokinetics, pharmacodynamics, toxicology, etc.
4) It was decided that in favor of Seven Dreamers Laboratories, Inc., Satoh Lab. in IIIS should be expanded to an endowed laboratory of clinical sleep medicine (2 faculty positions). Accordingly, Satoh steps away from the collaborative clinical research with Ibaraki Prefecture. We now look for his successor, who shall be appointed to Satellite PI to conduct the clinical research at Ibaraki Prefectural Medical Center of Psychiatry, which shall be our new Satellite. In addition, we appointed a researcher (Dr. Morita) in Forestry and Forest Products Research Institute in Tsukuba to a visiting associate professor of IIIS. Since she used to conduct the sleep analysis in a cohort research, she should manage the collaborative research of the genome cohort study (Nagahama Cohort Research) with Kyoto University.

In future, to give further solutions to the four agendas, the following initiatives are planned:

1. Improving and maintaining facilities and equipment to achieve a "World Premier International" standard.
   - Expanding the breeding capacity of the animal facility (ARC Satellite)
     The breeding facility located on 6th floor in ARC Satellite has been planned to have the max. capacity of 6,000 IVC, but only 50% of planned IVC racks and cages are currently installed. In future, the numbers of racks and cages will be increased and the breeding capability reinforced in accordance with research development.
   - Developing a clinical sleep lab
     A human metabolic chamber and one bed for clinical research will be installed in the clinical sleep lab located on the east side of the 3rd floor of the new research building.
   - Implementation of the future expansion space (the south side of the 4th floor)
     In the next few years, we aim at implementation of the future expansion space in the new research building, located on the south side of the 4th floor. Potential usages include an open-innovation drug discovery lab sponsored by pharmaceutical companies and hosting a research group for the JST basic research programs.
2. Further developing strength of IIIS such as capabilities of neuroscience in basic biological.
   Following Vice Center Director Sakurai's personnel shift, the staff members and a portion of the equipment in the current Sakurai Lab in Kanazawa University shall be transferred to IIIS. The transfer will enrich and strengthen the capabilities of Sakurai/Sakaguchi Lab significantly.
3. Filling gaps in research capabilities in pharmacokinetics, toxicology, etc., in pharmaceutical science.

Even in pharmaceutical departments in domestic and foreign universities, no labs could offer the research capabilities necessary for conducting collaboration of pre-clinical development of lead compounds, i.e., pharmacokinetics, pharmacodynamics and toxicology. It is thus necessary to consider continuously the collaboration with pharmaceutical companies to establish the "sleep science" by filling gaps in the research capabilities. In the research alliance with pharmaceutical companies, we will seek the best partner according to seeds/leads we could offer.

4. Strengthening capabilities in clinical sleep physiology, human molecular genetics, etc., in experimental medicine.

   Hereafter, to strengthen and expand research capabilities in experimental medicine, the endowed laboratory of clinical sleep medicine shall take the initiative along with the following joint research/collaboration with the satellites and external research institutes for additional translational research and clinical studies:

   a. Department of Neuropsychiatry, Akita University Graduate School of Medicine, and Center for Genomic Medicine, Kyoto University Graduate School of Medicine: molecular genetics of short sleepers and sleep disorders.
   b. Faculty of Health and Sport Science, University of Tsukuba: Effects of the orexin antagonist on cognitive and physical functions, and sleeping problems and solutions for athletes.
   c. JAXA Space Biomedical Research Office: Closed-environment stress and insomnia
   d. Ibaraki Prefectural Medical Center of Psychiatry: Sleep apnea syndrome and its counter-measure.
   e. Center for Cybernics Research, and Center for Computational Sciences, University of Tsukuba: Developing a diagnosis system for low-cost and accurate sleep measurement at home.

6-2. Measures to sustain the center as a World Premier International Research Center after Program Funding Ends

In the application for WPI program, University of Tsukuba committed itself to maintain IIIS as a permanent organization of the university after the end of the program implementation period. The University President and Vice President for Research have been confirming this commitment at every program committee meeting. As specific measures, the Vice President for Research and Vice President for Personnel Affairs are discussing how to qualify PI to be offered a tenure position at the end of the program.

Furthermore, in the third mid-term strategy plan starting from FY2016, University of Tsukuba will strive to establish a leading global research center or a "WPI-like" research center within the university. The university specifically set a goal for the third mid-term plan to evolve the TARA Center and the Center for Computational Sciences into the leading global research centers like IIIS positioned as the pioneer. The university has set IIIS as its flagship for the research centers and to sustain IIIS is its important task.

However, when this program's implementation period ends, the WPI subsidy more than 500 million yen/year will be also terminated. It would be impossible for the university to shoulder the full amount of funding under its current financial condition. Accordingly, downsizing the institute to some extent would be inevitable, and yet it is crucial to secure alternative funding.

At the completion of the FIRST program, external competitive research funds of IIIS had severely declined, but they are now recovering thanks to continuous efforts made by the Center Director, PIs and the Administrative Director in the last and current fiscal years. Hereafter, efforts will be made to obtain large competitive funds such as Grants-in-Aid for Scientific Research for Specially Promoted Research, Grants-in-Aid for Scientific Research on Innovative Areas, and Core Research for Evolutional Science and Technology (CREST) by JST. In addition, collaborative research with pharmaceutical companies and other companies focusing on products to improve sleep quality should be implemented to ensure sufficient external research funds. Currently 71% of the WPI subsidy are spent on labor costs, and we should improve the financial structure by shifting the fixed costs including the personnel expenditure covering postdocs and technicians from the WPI subsidy to external competitive funds.

As noted on 6-1, we will be attracting open-innovation drug discovery lab sponsored by pharmaceutical companies or hosting a research group for the JST Basic Research programs to the future expansion space in the new research building, which is located on the south side of the fourth floor, as a part of our efforts to obtain major research funds or grants.

As mentioned earlier, the research objectives for IIIS are 1) To elucidate the fundamental mechanisms of sleep/wake regulation, 2) To elucidate molecular pathogenesis of sleep disorders and

related diseases and 3) To develop treatments for sleep disorders. The third objective implies translating the newly developed sleep disorder treatments into practical applications. Provided that the collaboration with a global pharmaceutical company is successfully completed and the license revenues (the upfront payment, milestones, royalty) are partially fed back to IIIS, the income could cover the operational expenses of IIIS partially and help to sustain IIIS after the program funding ends.

The strategy and system to feed back or reinvest a part of the licensing revenues of WPI-funded research would be thus crucial to maintain IIIS as a "World premier international research center." Feasibility of ideas of the reinvesting system is being discussed with the university administration.

7. Others (within 1 page)

* In addition to the above 1-6 evaluation items, only if there is anything else that deserves mention regarding the center project's progress, please note it.

Inviting the new Vice Center Director

One important initiative made for an improvement in the organization management in FY2015 was the invitation of Sakurai from Kanazawa University to IIIS as Vice Center Director. Appointed as Vice Center Director on April 1, 2016, he would play a critical role in reinforcing IIIS's research capabilities in neuroscience as well as the operational management, and improving the guidance for young researchers. To provide him with a tenure position, he is appointed concurrently as a professor of IIIS and Faculty of Medicine.

New Collaborative PIs

The Collaborative PIs invited to IIIS from within the university are engaged in research collaboration varying in scope from PIs in the core group. Professor Junichi Hayashi, a Collaborative PI has retired on reaching retirement age in FY2014, and Professor Kumpei Tokuyama has joined as a new Collaborative PI since FY2015. Since Tokuyama has been performing for a long time in close collaboration with Professor Satoh, who was invited to the core group due to joint project with Ibaraki Prefecture, Professor Tokuyama is positioned as a joint PI for the Satoh Lab.

Foundation of an endowed laboratory of clinical sleep medicine

In FY2016, the Satoh Lab founded by the special collaborative research with Ibaraki Prefecture will be expanding as an endowed laboratory with the funds from the Seven Dreamers Laboratories Inc. This is the second case in which an endowed laboratory has been established within University of Tsukuba since the program started 17 years ago. Professor Satoh will be in charge of the endowed laboratory and another faculty will be recruited from the public. The collaboration with Tokuyama will continue. Ibaraki prefecture has agreed that Ibaraki Prefectural Medical Center of Psychiatry will become an IIIS Satellite for the special collaborative research with Ibaraki Prefecture, and the medical specialist in clinical hypnology should be recruited as a successor to Satoh and appointed to a Satellite PI. A job seminar for a candidate for the Satellite PI is scheduled in April.

8. Center's Response to Results of FY2015 Follow-up (including Site Visit Results) (Use as Many Pages as needed.)

Describe the Center's Response to Results of FY2015 Follow-up. Note: If you have already provided this information, please indicate where in the report.

Remark 1: The result of mutant mice and genes identified by forward genetics approach must be published in research papers before the next site visit for interim assessment.

Response: There has been significant progress in the functional analysis and characterization of the novel genes regulating sleep/wake. We have already submitted the papers as described in 2-1.

Remark 2: Both Professor Yanagisawa and Funato should explain the strategies to elucidate the functional analysis of the sleepy and dreamless gene's (signal transduction, expression profile etc.).

Response: The strategies of the functional analysis have been already implemented as explained above and described in 2-1.

Remark 3: The reduction of external research funds for FY2015 remains a major concern and must be improved for FY2016 and onward. All PIs must obtain large research funds. It should be supported for the PIs, who will be applying for external grants in languages other than Japanese.

Response: External research funds obtained in FY2015 roughly doubled compared to that in FY2014.

Although it is still to be increased, our financial conditions were improved significantly. We will continue the efforts to obtain more competitive research funds, not only from JSPS, JST, MEXT, MHLW and MAFF, but also from METI, AMED, etc., and further improvements are expected for FY2016 as described in 2-6.

Remark 4: IIIS should describe how to bridge the basic research results using animals into clinical research in humans through the collaboration with clinicians or other human research teams.

Response: We consider the translational research bridging achievements in basic biology to experimental medicine or clinical research is crucial to achieve our objectives. As described 2-2, to enforce the translational research, our major efforts have been dedicated to increase and expand collaboration/research alliances with outside groups including groups in University of Tsukuba, the satellites, external research institutions, and even research groups in industries, as our new challenges.

Remark 5: The inclusion of female PI employment in the core group is crucial. Recruitment must be continued, however few candidates there are, along with developing the female researchers into the junior PI level.

Response: As for female PI employment in the core group, we have actively been recruiting with female only PI advertisements on a number of websites and journals since October 2014. We have already conducted job seminars for 3 candidates. At present, there have been no successful candidates; however, we will continue our recruiting efforts. We also consider promotion of female researchers to junior PI level as described in 5-5-2.

Remark 6: An adequate mentoring system is necessary given soaring numbers of students. Please propose official plans.

Response: IIIS also considers mentoring crucial. University of Tsukuba separates educational organizations (undergraduate and graduate school) and research organizations (faculty organizations), and IIIS belongs in the latter group. Accordingly, when an IIIS researcher is to provide students with research guidance or mentoring, a teaching qualification must be approved with examinations by the educational organizations such as the graduate schools. Usually, PI gets the teaching qualification (research guidance) as a mentor, while other faculty members in his/her Lab assist as required. Other research centers in the university operate the same system, which is the standard within University of Tsukuba.

Specifically, within the IIIS core group PIs, 10 with Graduate School of Comprehensive Human Science Biomedical Sciences Majors of Medical Science (Ph.D. programs), 9 with Graduate School of Comprehensive Human Science Master's Program in Medical Sciences, 9 with the Ph.D. Program in Human Biology (HBP), and one with Doctoral Program in Chemistry, Graduate School of Pure and Applied Sciences Chemistry, have been qualified for teaching/mentoring as described in 5-5-1.

World Premier International Research Center Initiative (WPI)

Appendix 1-1. FY 2015 List of Principal Investigators

NOTE:
・Underline names of investigators who belong to an overseas research institution. Place an asterisk (*) by names of investigators considered to be ranked among world's top researchers.
・In case of researchers not listed in the latest report, attach "Biographical Sketch of a New Principal Investigator".

<Results at the end of FY2015>

Principal Investigators Total:22

Name (Age)	Affiliation (Position title, department, organization)	Academic degree, specialty	Working hours (Total working hours: 100%)				Starting date of project participation	Status of project participation (Describe in concrete terms)	Contributions by PIs from overseas research institutions
			Work on center project		Others				
			Research activities	Other activities	Research activities	Other activities			
Center director Masashi Yanagisawa (55)*	Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	M.D., Ph.D. Neuroscience, Pharmacology	75%	20%	4%	1%	December 2012	Usually stays at the center	
Takeshi Sakurai (51)*	Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	M.D., Ph.D., Neuroscience	10%	10%	50%	30%	April 2013	We are studying: a) Neural mechanisms that control sleep/wakefulness states b) Relationship between the sleep/wake control and emotion and emotional memory	
Hiromasa Funato (46)*	Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba Associate Professor, Univ Toho	M.D., Ph.D. Neuroscience	40%	5%	25%	30%	December 2012	Usually stays at the center three times a week	

Name (Age)	Affiliation (Position title, department, organization)	Academic degree, specialty	Working hours (Total working hours: 100%)				Starting date of project participation	Status of project participation (Describe in concrete terms)	Contributions by PIs from overseas research institutions
			Work on center project		Others				
			Research activities	Other activities	Research activities	Other activities			
Yoshihiro Urade (62)*	Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	Ph.D. Biochemistry Neuroscience	45%	5%	45%	5%	October 2013	Usually stays at the center	
Robert Greene (65)*	Professor, Department of Psychiatry, University of Texas Southwestern Medical Center, Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	M.D., Ph.D. Neuroscience	10%	0%	70%	20%	December 2013	a) visit center 3X/yr for ~2 weeks /visit b) Skype meeting with lab 1X/week c) attend (by Skype) PI meeting 1X/month d) participate in person with the annual IIIS symposium e) participate in person in annual Site Visit	Collaboration of ongoing research project at UTSW investigating role of adenosine in homeostatic sleep control
Qinghua Liu (44)*	Associate Professor, Department of Biochemistry, University of Texas Southwestern Medical Center Professor, Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	Ph.D. Genetics, Molecular Biology, Biochemistry	33%	2%	60%	5%	April 2013	a) Stays at the center for 3 weeks every 2-3 months, total 3-4.5 months/year; b) Joins a videoconference from US >2 times a week	Accept young scientists to WPI center (10/period)
Hiroshi Nagase (68)*	Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	Ph.D. Medicinal Chemistry, Organic Chemistry	65%	0%	30%	5%	April 2013	Usually stays at the center	

Name (Age)	Affiliation (Position title, department, organization)	Academic degree, specialty	Working hours (Total working hours: 100%)				Starting date of project participation	Status of project participation (Describe in concrete terms)	Contributions by PIs from overseas research institutions
			Work on center project		Others				
			Research activities	Other activities	Research activities	Other activities			
Makoto Satoh (60)	Center director, Sleep Medical Center, Ibaraki Prefectural Medical Center of Psychiatry Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	M.D., Sleep Medicine	10%	5%	15%	70%	April 2015	Stays at the center once a week	
Ichiyo Matsuzaki (56)*	Professor, Faculty of Medicine, University of Tsukuba	M.D., Ph.D. Occupational Psychiatric Medicine, Space Medicine	5%	5%	60%	30%	March 2013	About 10% of effort. The remaining is allocated for Faculty of Medicine.	
Hitoshi Shimano (56)*	Professor, Faculty of Medicine, University of Tsukuba	M.D., Ph.D. Endocrinology, Metabolism	10%	5%	55%	30%	March 2013	Attempts to elucidate molecular mechanisms of links between endocrinological/metabolic disturbances and sleep disturbances	
Kumpei Tokuyama (62)	Professor, Faculty of Health and Sport Sciences, University of Tsukuba	Ph.D., Sports Medicine	20%	0%	40%	40%	April 2015	Stays at the center once a week	

Name (Age)	Affiliation (Position title, department, organization)	Academic degree, specialty	Working hours (Total working hours: 100%)				Starting date of project participation	Status of project participation (Describe in concrete terms)	Contributions by PIs from overseas research institutions
			Work on center project		Others				
			Research activities	Other activities	Research activities	Other activities			
Akiyoshi Fukamizu (56)*	Professor, Tsukuba Advanced Research Alliance, University of Tsukuba	Ph.D., Molecular Biology	1%	1%	50%	48%	March 2013	Started the collaboration with Chika Miyoshi (Yanagisawa/Funato Lab.).	
Satoru Takahashi (54)*	Professor, Laboratory Animal Resource Center, Department of Anatomy and Embryology, Faculty of Medicine, University of Tsukuba	M.D., Ph.D. Developmental biology	10 %	10 %	40 %	40 %	March 2013	Participate in generation of genetically modified mice by using CRISPR/Cas9 system at Laboratory Animal Resource Center	
Joseph Takahashi (64)*	Professor, Department of Neuroscience, University of Texas Southwestern Medical Center	Ph.D., Neuroscience	5%	0%	75%	20%	December 2012	Collaborator on ENU mutagenesis screen for sleep mutants, and genetic mapping; Collaborator on the establishing the role of SCN NMS-producing neurons in circadian rhythms	Collaboration. Available to accept young scientists from WPI for collaborative projects.
Carla Green (53)*	Professor, Department of Neuroscience, University of Texas Southwestern Medical Center	Ph.D. Molecular Biology, Biochemistry, Circadian rhythms	2%	3%	90%	5%	March 2013	We have established the TAIL-seq procedure in our lab and are currently testing it for efficacy in measuring polyA tail length and polyadenylation site usage. We will then proceed with our plans to measure these parameters in brain tissue from mice that have undergone sleep deprivation and recovery sleep.	Attended two IIIS symposia (as speaker and session chair) as well as the external review committee meeting in 2015
Yang Dan (48)*	Professor, Department of Molecular and Cell Biology, University of California, Berkeley, HHMI	Ph.D., Neurobiology	3%	2%	85%	10%	April 2014	Usually stays at the satellite center	

Name (Age)	Affiliation (Position title, department, organization)	Academic degree, specialty	Working hours (Total working hours: 100%)				Starting date of project participation	Status of project participation (Describe in concrete terms)	Contributions by PIs from overseas research institutions
			Work on center project		Others				
			Research activities	Other activities	Research activities	Other activities			
Tetsuo Shimizu (63)*	Professor, Department of Neuropsychiatry, Akita University Graduate School of Medicine	M.D., Ph.D., Psychiatry	10%	5%	20%	65%	April 2013	Joins a video conference from Akita University once a month	
Hitoshi Okamura (63)*	Professor, Graduate School of Pharmaceutical Sciences, Kyoto University Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	M.D., Ph.D. Chronobiology	3%	0%	67%	30%	July 2015	Usually stays at the satellite center	
Kaspar Vogt (49)	Associate Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	M.D., Ph.D. Physiology, Pharmacology, Neurobiology	80%	20%	0%	0%	February 2014	Usually stays at the center	
Michael Lazarus (46)	Associate Professor International Institute for Integrative Sleep Medicine, University of Tsukuba	Ph.D. Neuroscience	95%	5%	0%	0%	April 2013	Usually stays at the center	
Masanori Sakaguchi (39)	Associate Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	M.D., Ph.D. Brain regeneration and sleep	95%	5%	0%	0%	February 2013	Usually stays at the center	

Name (Age)	Affiliation (Position title, department, organization)	Academic degree, specialty	Working hours (Total working hours: 100%)				Starting date of project participation	Status of project participation (Describe in concrete terms)	Contributions by PIs from overseas research institutions
			Work on center project		Others				
			Research activities	Other activities	Research activities	Other activities			
Yu Hayashi (35)	Assistant professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	Ph.D. Science	70%	10%	10%	10%	April 2013	Usually stays at the center	

Researchers unable to participate in project in FY 2015

Name	Affiliation (Position title, department, organization)	Starting date of project participation	Reasons	Measures taken
Junichi Hayashi	Professor, Faculty of Life and Environmental Sciences, University of Tsukuba	March 2013	Mandatory retirement	Makoto Satoh and Kumpei Tokuyama newly joined
Natsuko Kanda	Assistant Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba	April 2013	Retirement for personal reasons	
Deependra Kumar	Researcher, International Institute for Integrative Sleep Medicine, University of Tsukuba	September 2014	Selected as a JSPS Postdoctoral Fellow for Overseas Researchers (Under the 2nd recruitment of FY 2015-2016 JSPS Postdoctoral Fellowship Program)	Continuously stays at the center
Tomoyuki Fujiyama	Researcher, International Institute for Integrative Sleep Medicine, University of Tsukuba	April 2014	Selected as a JSPS Research Fellow for Young Scientists (Under the JSPS Research Fellowship for Young Scientists Program)	Continuously stays at the center

World Premier International Research Center Initiative (WPI)

Biographical Sketch of a New Principal Investigator

Name (Age) *NOTE: Place an asterisk (*) by the name of investigators considered to be ranked among the world's top researchers.*	Hitoshi Okamura (63)*
Current affiliation (Position title, department, organization)	Professor, Graduate School of Pharmaceutical Sciences, Kyoto University Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba
Academic degree, specialty	M.D., Ph.D. Chronobiology

Research and education history

-1979 Kyoto Prefectural University of Medicine (M.D., Ph.D.)
1979-1981 Medical intern, Children's Medical Center, Okayama National Hospital
1981-1987 Assistant, Department of Anatomy II, Kyoto Prefectural University of Medicine
1987-1990 Lecturer, Department of Anatomy II, Kyoto Prefectural University of Medicine
1987-1989 Internship, INSERM (Lyon) and CNRS (Gif-sur-Yvette)
1990-1995 Associate Professor, Department of Anatomy II, Kyoto Prefectural University of Medicine
1995-2000 Professor, Department of Anatomy II, Kobe University School of Medicine
2000-2008 Professor, Division of Molecular Brain Science, Kobe University Graduate School of Medicine
2007- Professor, Systems Biology, Kyoto University Graduate School of Pharmaceutical Sciences
2014- Research Director, The Japan Science Technology Institute, CREST.
2015- Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba

Achievements and highlights of past research activities

Hitoshi Okamura is a researcher studying the molecular mechanisms of mammalian circadian clock. Past achievements of Dr. Okamura and his team include the identification of mammalian *Period* genes, measurements of circadian oscillations in a single cell, and the demonstration that the cell cycle is under clock control. He also pioneered the idea that interneuronal signaling is indispensable for the generation of the circadian oscillations in the suprachiasmatic nucleus (SCN), the master clock in mammals. Recently, his team demonstrated the importance of GPCR and circadian regulation of downstream signaling in the SCN, and identified a molecular mechanism underlying jet lag. Furthermore, they demonstrated that RNA methylation-dependent mRNA processing is required for accurate timing, and discovered a novel steroidogenic gene causing salt-induced hypertension when the clock is disrupted.

Achievements

(1) International influence *a) Guest speaker, chair, director, or honorary member of a major international academic society in the subject field, b) Holder of a prestigious lectureship, c) Member of a scholarly academy in a major country, d) Recipient of an international award(s) , e) Editor of an influential journal etc.*

a) Guest speaker: 1. Okamura H: New inter- and intracellular regulations of the circadian pacemaker, in 14th SRBR meeting, June 15-18, 2014, at Big Sky, Montana (USA). 2. Okamura H: Clock genes and diseases, in Keystone Symposium on Molecular Clockworks and the Regulation of Cardio-Metabolic Function, April 3-7, 2013, Snowbird (USA) 3. Okamura H: (Plenary Lecture) Clock and hypertension. 15th International Congress of Endocrinology & 14th ECE, May 05-09, 2012 Florence (Italy). 4. Okamura H: (Invited Speaker) Clock Genes and Hypertension. The 94th Annual Meeting of The Endocrine Society. (ENDO 2012), June 23-26, 2012, Houston (USA). 5. Okamura H: Clock gene, aldosterone and hypertension, in 24th Meeting of the International Society of Hypertension (ISH) Hypertension Sydney 2012, Sept 30 - Oct 4, 2012, Sydney (Australia) and others. Invited as a guest speaker to 47 overseas international scientific conferences, to 20 universities overseas and to 39 domestic international scientific conferences in total.

(2) Receipt of large-scale competitive funding *(over past 5 years)*

2014-2019 “Chronometabolism: Molecular analysis of biological timing” on CREST, the Strategic Basic Research Programs by JST (Japan Science and Technology Agency), ¥366,900,000-, Research Director

(3) Article citations *(Titles of major publications, and number of citations.)*

1. Matsuo T, Yamaguchi S, Mitsui S, Emi A, Shimoda F, Okamura H: Control mechanism of the circadian clock for timing of cell division in vivo. Science 302: 255-259, 2003. Citations: 574
2. Shigeyoshi Y, Taguchi K, Yamamoto S, Takekida S, Yan L, Tei H, Moriya T, Shibata S, Loros JJ, Dunlap JC, Okamura H: Light-induced resetting of a mammalian circadian clock is associated with rapid induction of the mPer1 transcript. Cell 91: 1043-1053, 1997. Citations: 570
3. Tei H, Okamura H, Shigeyoshi Y, Fukuhara C, Ozawa R, Hirose M, Sakaki Y: Circadian oscillation of a mammalian homologue of the Drosophila period gene. Nature 389, 512-516, 1997. Citations: 552
4. Yamaguchi S, Isejima H, Matsuo T, Okura R, Yagita K, Kobayashi M, Okamura H: Synchronization of cellular clocks in the suprachiasmatic nucleus. Science 302: 1408-1412, 2003. Citations: 460

(4) Others *(Other achievements that indicate qualification as a top-caliber researcher, if any.)*

Awards:
1998 Nakaakira Tsukahara Memorial Award
2001 Inoue Prize for Science
2003 Medical Award of The Japan Medical Association
2004 Iue Cultural Prize
2007 Purple Ribbon Medal
2009 Aschoff-Ruler Prize
2016 TORAY Science and Technology Award

World Premier International Research Center Initiative (WPI)

Biographical Sketch of a New Principal Investigator

Name (Age) *NOTE: Place an asterisk (*) by the name of investigators considered to be ranked among the world's top researchers.*	Makoto Satoh (60)
Current affiliation (Position title, department, organization)	Principal Investigator and Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba
Academic degree, specialty	M.D, Ph.D., Sleep Medicine, Environmental Physiology

Research education history
1987 Research Fellow, Tohoku University
1989 Research Fellow, Niigata University
1996 Postdoctoral Fellow, University of Wisconsin, Madison
1998 Postdoctoral Fellow, Niigata University
2001 Professor, Joetsu University of Education,
Director, University Health Center
2005 Professor, University of Tsukuba
Director, Center of Sleep Disordered Breathing, University of Tsukuba Hospital
2015 Director, Ibaraki Prefectural Center for Sleep Medicine and Sciences
2015 Principal Investigator and Professor, International Institute for Integrative Sleep Medicine, University of Tsukuba

Education
1982 Niigata University School of Medicine (MD)
1991 Niigata University School of Medicine (Ph.D.)

Achievements and highlights of past research activities *(Describe qualifications as a top-caliber researcher if he/she is considered to be ranked among the world's top researchers.)*

I have been a clinician specialized in sleep apnea and other sleep disorders. I invented Nasal Airway Stent (NAS), which is a new treatment tool for obstructive sleep apnea. The NAS is expected alternative treatment for patients with OSA and may be used as an immediate therapeutic tool while the patient loses weight or as an alternative for those patients who fail or cannot tolerate nasal CPAP.

Achievements

(1) International influence *a) Guest speaker, chair, director, or honorary member of a major international academic society in the subject field, b) Holder of a prestigious lectureship, c) Member of a scholarly academy in a major country, d) Recipient of an international award(s) , e) Editor of an influential journal etc.*

(2) Receipt of large-scale competitive funding *(over past 5 years)*

(3) Article citations *(Titles of major publications, and number of citations.)*

Selected articles

- Iwayama K, Kurihara R, Nabekura Y, Kawabuchi R, Park I, Kobayashi M, Ogata H, Kayaba M, Satoh M, Tokuyama K. Exercise Increases 24-h Fat Oxidation Only When It Is Performed Before Breakfast. *EBioMedicine* **2**(12): 2003–2009, 2015 (1 citation)
- Ito Y, Takahashi S, Shen M, Yamaguchi K, Satoh M. Effects of L-serine ingestion on human sleep. *Springerplus* **3**:456, 2014 (1 citation)
- Yajima K, Seya T, Iwayama K, Hibi M, Hari S, Nakashima Y, Ogata H, Omi N, Satoh M, Tokuyama K. Effects of Nutrient Composition of Dinner on Sleep Architecture and Energy Metabolism during Sleep. *J Nutr Sci*

Vitaminol **60**(2):114-21, 2014 (0 citation)

- Hosono H, Homma M, Satoh M, Kohda Y. Variables influencing patient satisfaction for hypnotics: Difference between zolpidem and brotizolam. *J. Clin. Pharm. Ther*. **39**(5):507-10, 2014 (1 citation)
- Kayaba M, Iwayama K, Ogata H, Seya Y, Tokuyama K, Satoh M. Drowsiness and low energy metabolism in the following morning induced by nocturnal blue light exposure. *Sleep Med* **14**(1): e166-e167, 2013 (0 citation)
- Naruse Y, Tada H, Satoh M, Yanagihara M, Tsuneoka H, Hirata Y, Ito Y, Kuroki K, Machino T, Yamasaki H, Igarashi M, Sekiguchi Y, Sato A, Aonuma K. Concomitant obstructive sleep apnea increases the recurrence of atrial fibrillation following radiofrequency catheter ablation of atrial fibrillation: Clinical impact of continuous positive airway pressure therapy. *Heart Rhythm* **10**(3): 331-7, 2013 (0 citation)
- Naruse Y, Tada H, Satoh M, Yanagihara M, Tsuneoka H, Hirata Y, Machino T, Yamasaki H, Igarashi M, Kuroki K, Ito Y, Sekiguchi Y, Aonuma K. Radiofrequency catheter ablation of persistent atrial fibrillation decreases a sleep-disordered breathing parameter during a short follow-up period. *Circ J.* **76**(9):2096-103, 2012 (3 citations)
- Koshino Y, Satoh M, Katayose Y, Yasuda K, Yamasaki H, Sekiguchi Y, Kuga K, Aonuma K. Nocturnal premature ventricular complexes in a young woman with respiratory effort-related arousals. *Sleep and Biological Rhythms* **8**(1): 79–82, 2010 (0 citation)
- Koshino Y, Satoh M, Katayose Y, Kuroki K, Sekiguchi Y, Yamasaki H, Yoshida K, Yasuda K, Tanigawa T, Kuga K, Aonuma K. Sleep apnea and ventricular arrhythmias: Clinical outcome, electrophysiologic characteristics, and follow-up after catheter ablation. *J Cardiol.* **55**(2):211-6, 2010 (12 citations)
- Katayose Y, Tasaki M, Ogata H, Nakata Y, Tokuyama K, Satoh M. Metabolic rate and fuel utilization during sleep assessed by whole-body indirect calorimetry. *Metabolism* **58**(7):920-6, 2009 (30 citations)
- Koshino Y, Satoh M, Katayose Y, Yasuda K, Tanigawa T, Takeyasu N, Watanabe S, Yamaguchi I, Aonuma K. Association of Sleep-Disordered Breathing and Ventricular Arrhythmias in Patients without Heart Failure. *Am J Cardiol.* **101**(6):882-6, 2008 (27 citations)
- Sakai K, Takada T, Nakayama H, Kubota Y, Nakamata M, Satoh M, Suzuki E, Akazawa K, Gejyo F. Serotonin-2A and 2C receptor gene polymorphisms in Japanese patients with obstructive sleep apnea. *Intern Med*. **44**(9):928-33, 2005 (43 citations)
- Kubota Y, Nakayama H, Takada T, Matsuyama N, Sakai K, Yoshizawa H, Nakamata M, Satoh M, Akazawa K, Suzuki E, Gejyo F. Facial axis angle as a risk factor for obstructive sleep apnea. *Intern Med*. **44**(8):805-10, 2005 (28 citations)
- Kinebuchi S, Kazama JJ, Satoh M, Sakai K, Nakayama H, Yoshizawa H, Narita I, Suzuki E, Gejyo F. Short-term use of continuous positive airway pressure ameliorates glomerular hyperfiltration in patients with obstructive sleep apnoea syndrome. *Clin Sci* (Lond). **107**(3):317-22, 2004 (56 citations)
- Hida W, Okabe S, Tatsumi K, Kimura H, Akasiba T, Chin K, Ohi M, Nakayama H, Satoh M, Kuriyama T. Nasal continuous positive airway pressure improves quality of life in obesity hypoventilation syndrome. *Sleep Breath* **7**(1):3-12. 2003 (49 citations)
- Nakano M, Hasegawa H, Takada T, Ito S, Muramatsu Y, Satoh M, Suzuki E, Gejyo F. Pulmonary diffusion capacity in patients with systemic lupus erythematosus. *Respirology* **7**(1):45-9, 2002 (24 citations)
- Satoh M, Eastwood PR, Smith CA, Dempsey JA. Nonchemical elimination of inspiratory motor output via mechanical ventilation in sleep. *Am J Respir Crit Care Med*. **163**(6):1356-64, 2001 (23 citations)

(4) Others *(Other achievements that indicate qualification as a top-caliber researcher, if any.)*

World Premier International Research Center Initiative (WPI)

Biographical Sketch of a New Principal Investigator

Name (Age) *NOTE: Place an asterisk (*) by the name of investigators considered to be ranked among the world's top researchers.*	Kumpei Tokuyama (62)
Current affiliation (Position title, department, organization)	Professor, Faculty of Health and Sport Sciences, University of Tsukuba
Academic degree, specialty	Ph.D., Sports Medicine

Research

1983	Postdoctoral Fellow, University of Southern California
1984	Postdoctoral Fellow, Ottawa University
1986	Postdoctoral Fellow, Swarthmore College
1988	Associate Professor, Osaka Shoin Womens' College
1992	Lecturer, University of Tsukuba
1997	Associate Professor, University of Tsukuba
2007	Professor, University of Tsukuba

Education

1976	University of Tokyo Education (B.P.Ed.)
1979	Tsukuba University (M.P.Ed.)
1983	Ehime University School of Medicine (Ph.D.)

Achievements and highlights of past research activities *(Describe qualifications as a top-caliber researcher if he/she is considered to be ranked among the world's top researchers.)*

I improved the time response of whole room indirect calorimeter, time resolution of which is currently the best in the world, which enabled us to find differences in energy metabolism among sleep stages.

Achievements

(1) International influence *a) Guest speaker, chair, director, or honorary member of a major international academic society in the subject field, b) Holder of a prestigious lectureship, c) Member of a scholarly academy in a major country, d) Recipient of an international award(s) , e) Editor of an influential journal etc.*

Keynote Lecture:
Recent Advances and Controversies in Measuring Energy Metabolism, Tokyo, 2011
Ergonic Aids and Nutritional Supplements for Health and Sports, Thailand, 2016

(2) Receipt of large-scale competitive funding *(over past 5 years)*

Sports Research Innovation Project（Japan Sports Agency, ¥45,000,000／yr, 2015-2020, member of research project)

(3) Article citations *(Titles of major publications, and number of citations.)*

Selected articles

- Obata A, Kubota N, Kubota T, Iwamoto M, Sato H, Sakurai Y, Takamoto I, Katsuyama H, Suzuki Y, Tokuyama K, *et al*. Tofogliflozin Improves Insulin Resistance in Skeletal Muscle and Accelerates Lipolysis in Adipose Tissue in Male Mice. *Endocrinology* **157**(3):1029-42, 2015 (1 citation)
- Iwayama K, Kurihara R, Nabekura Y, Kawabuchi R, Park I, Kobayashi M, Ogata H, Kayaba M, Satoh M, Tokuyama K. Exercise Increases 24-h Fat Oxidation Only When It Is Performed Before Breakfast. *EBioMedicine* **2**(12): 2003–2009, 2015 (1 citation)
- Yajima K, Seya T, Iwayama K, Hibi M, Hari S, Nakashima Y, Ogata H, Omi N, Satoh M, Tokuyama K. Effects of Nutrient Composition of Dinner on Sleep Architecture and Energy Metabolism during Sleep. *J Nutr Sci Vitaminol* **60**(2):114-21, 2014 (0 citation)
- Lan F, Misu H, Chikamoto K, Takayama H, Kikuchi A, Mohri K, Takata N, Hayashi H, Matsuzawa-Nagata N, Tokuyama K *et al*. LECT2 functions as a hepatokine that links obesity to

skeletal muscle insulin resistance. *Diabetes* **63**(5):1649-64, 2014 (7 citations)

- Okada-Iwabu M, Yamauchi T, Iwabu M, Honma T, Hamagami K, Matsuda K, Yamaguchi M, Tanabe H, Kimura-Someya T, Tokuyama K et al. A small-molecule AdipoR agonist for type 2 diabetes and short life in obesity. *Nature* **503**:493-399, 2013 (91 citations)
- Nakaya K, Kubota N, Takamoto I, Kubota T, Katsuyama H, Sato H, Tokuyama K, Hashimoto S, Goto M, Jomori T, *et al*. Dipeptidyl peptidase-4 inhibitor anagliptin ameliorates diabetes in mice with haploinsufficiency of glucokinase on a high-fat diet. *Metabolism* **62**(7):939-51, 2013 (7 citations)
- Iwabu M, Yamauchi T, Okada-Iwabu M, Sato K, Nakagawa T, Funata M, Yamaguchi M, Namiki S, Nakayama R, Tokuyama K *et al*. Adiponectin and adipoR1 regulate PGC-1α and mitochondria by Ca^{2+} and AMPK/SIRT1. *Nature* **464**:1313-9, 2010 (323 citations)
- Kubota N, Kubota T, Itoh S, Kumagai H, Kozono H, Takamoto I, Mineyama T, Ogata H, Tokuyama K, Ohsugi M *et al*. Dynamic Functional Relay between Insulin Receptor Substrate 1 and 2 in Hepatic Insulin Signaling during Fasting and Feeding. *Cell Metab*. **8**(1):49-64, 2008 (106 citations)
- Yamauchi T, Nio Y, Maki T, Kobayashi M, Takazawa T, Iwabu M, Okada-Iwabu M, Kawamoto S, Kubota N, Tokuyama K *et al*. Targeted disruption of AdipoR1 and AdipoR2 causes abrogation of adiponectin binding and metabolic actions. *Nat. Med*. **13**: 332-339, 2006 (633 citations)
- Kamei N, Tobe K, Suzuki R, Ohsugi M, Watanabe T, Kubota N, Ohtsuka-Kowatari N, Kumagai K, Sakamoto K, Tokuyama K *et al*. Overexpression of monocyte chemoattractant protein-1 in adipose tissues causes macrophage recruitment and insulin resistance. *J. Biol. Chem*. **281**: 26602-26614, 2006 (463 citations)

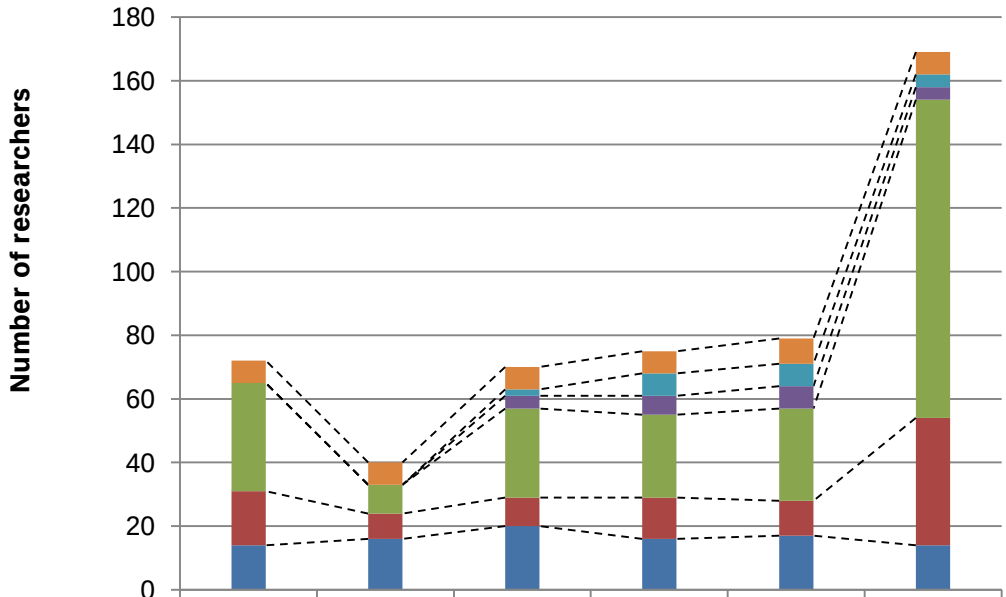
(4) Others *(Other achievements that indicate qualification as a top-caliber researcher, if any.)*

World Premier International Research Center Initiative (WPI)

Appendix 1-2. Number of researchers in the "core" established within the host institution

*Make a graph of the annual transition in the number of center personnel since the start of project.

Annual Transition in the Number of Members



	At beginning	FY2012	FY2013	FY2014	FY2015	Final goal
PI : Researchers from within host institution	7	7	7	7	8	7
PI : Foreign researchers invited from abroad	0	0	2	7	7	4
PI : Researchers invited from other Japanese institutions	0	0	4	6	7	4
Other researchers	34	9	28	26	29	100
Research support staffs	17	8	9	13	11	40
Administrative staffs	14	16	20	16	17	14

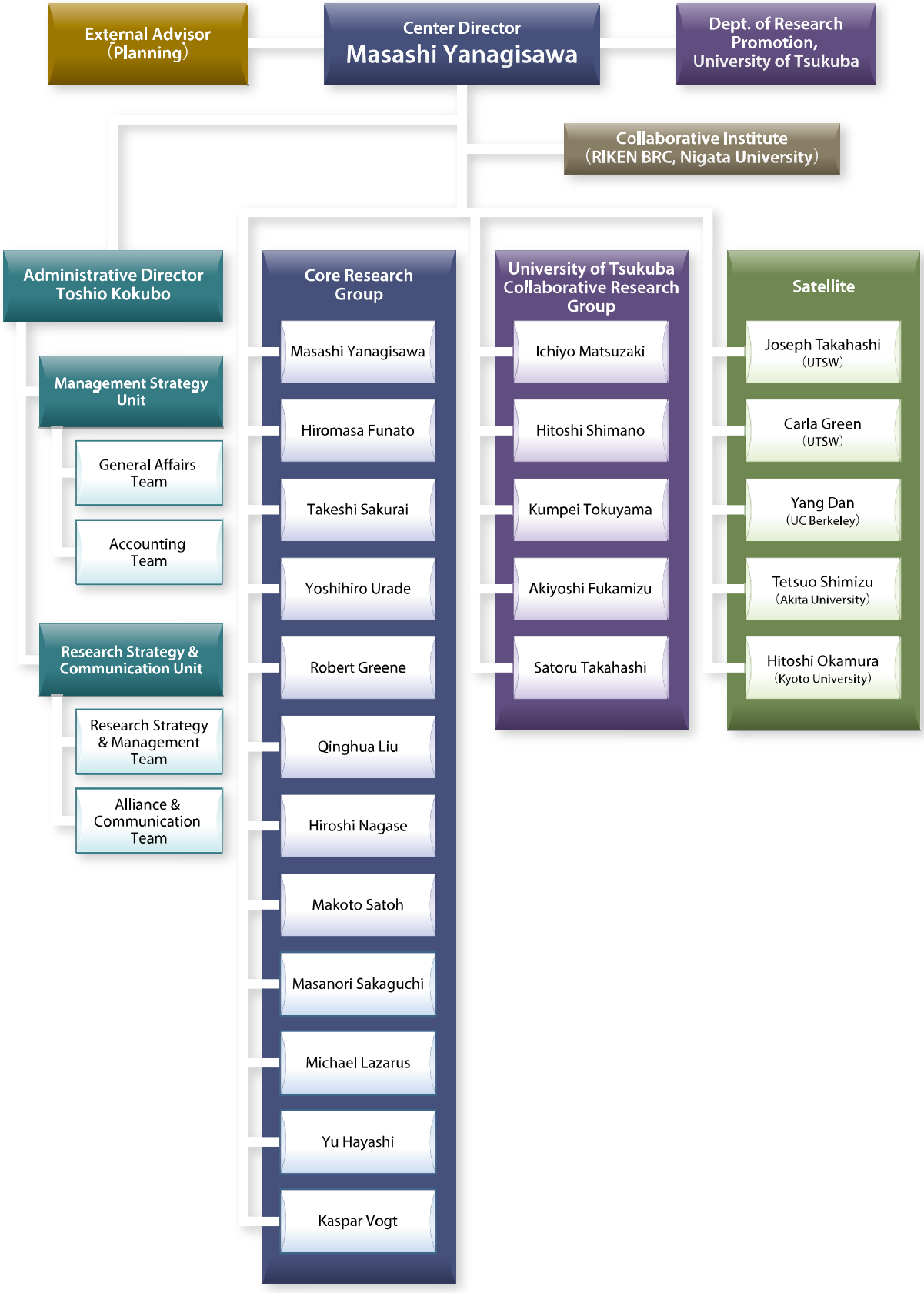
- Enter matters warranting special mention, such as concrete plans for achieving the Center's goals, established schedules for employing main researchers, particularly principal investigators.

To hire female PIs and young researchers including postdoctoral fellows into core groups, IIIS engages in international open recruitment by placing job advertisements on various websites. The number of applicants are gradually increasing. We successfully recruited PIs and postdocs utilizing opportunities via the international network of core and satellite PIs, thus we continue this effort to further recruit top caliber researchers to achieve the goal.

World Premier International Research Center Initiative (WPI)

Appendix 1-3. Center's Management System

- Please diagram management system in an easily understood manner.



World Premier International Research Center Initiative (WPI)

Appendix 1-4. Campus Map

– Please draw a simple map of the campus showing where the main office and principle investigator(s) are located.

IIIS Building

5/6F Animal Facility (ARC Satellite)

4F + Future Expansion Space


Nagase

3F


Sakurai


Sakaguchi


Greene


Vogt


Urade


Lazarus


Satoh

2F


Yanagisawa

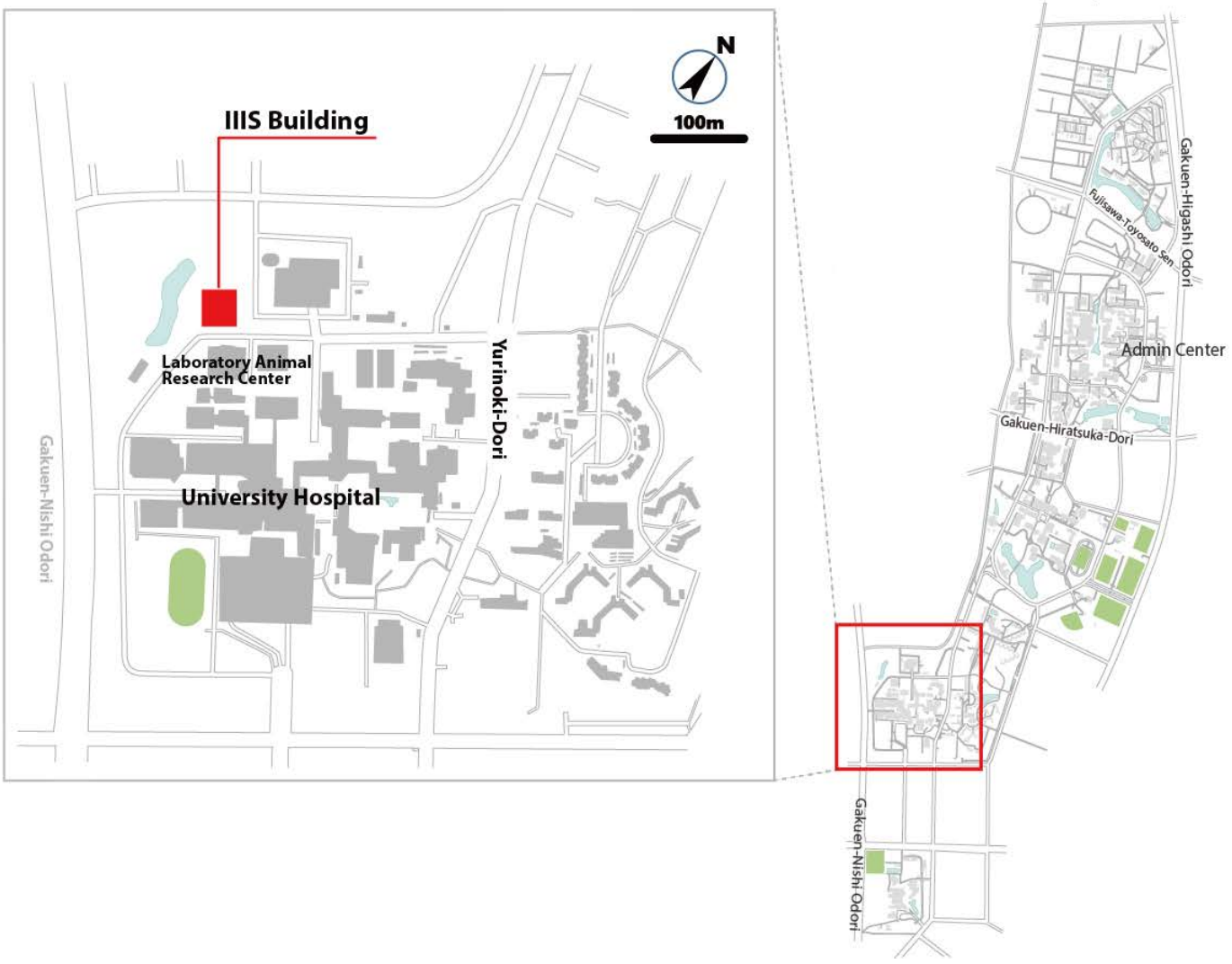

Funato


Liu


Hayashi

1F Admin Office, Auditorium, Hall, Lounge

Campus Map



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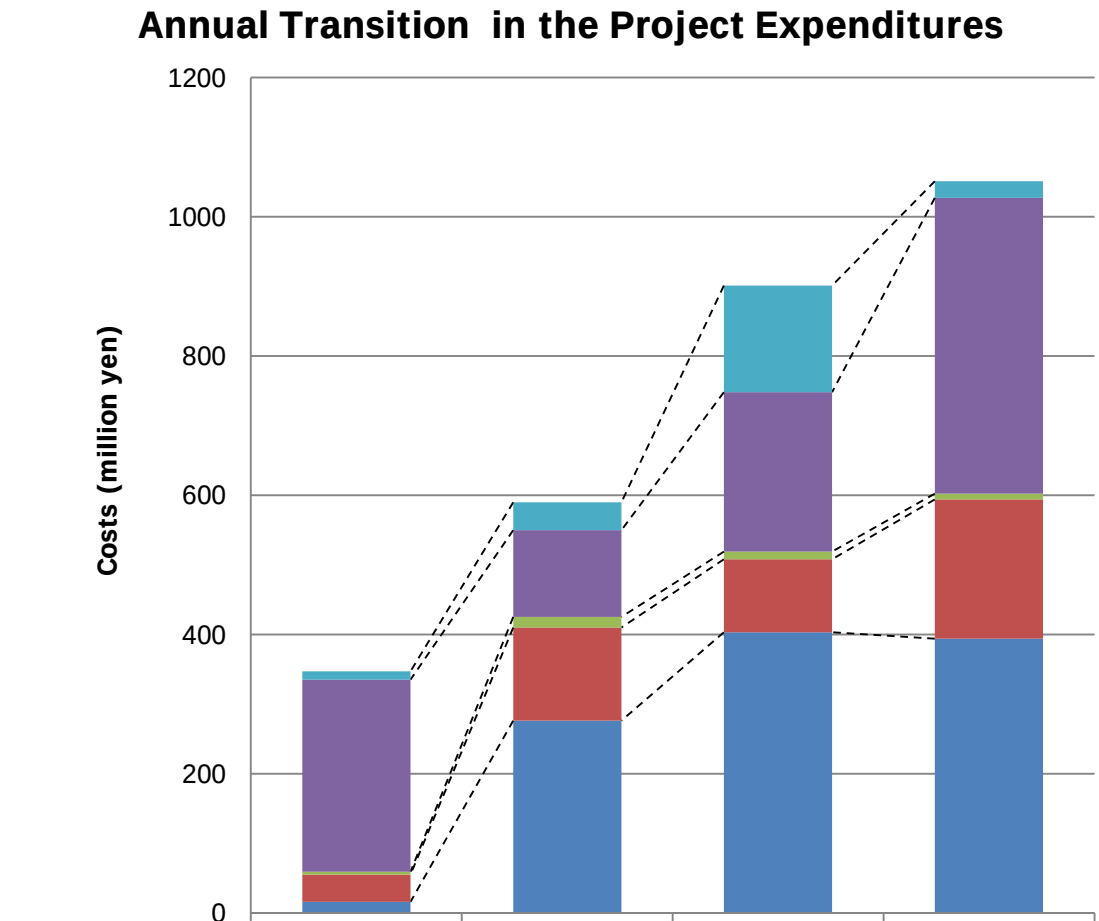
Appendix 1-5-1. Annual transition in the project expenditures

*Make a graph of the transition in the number of overall project funding.



	FY2012	FY2013	FY2014	FY2015
Research projects	12	40	153	24
Equipment	276	125	229	425
Travel	4	15	11	8
Project activities	39	134	105	200
Personnel	16	276	403	394

- To date, what has the Center's thinking been about spending project funding, and how has the funding been spent?

The project expenditures have grown rapidly mainly due to the increases in the item of "Equipment," which includes a major portion of depreciation of buildings and equipment. After subtracting the depreciation from the expenditures, the personnel expenses occupy about 70% of the rest. Since the labor costs are fixed costs, we should reduce them by shifting a part of them from the project expenses to external competitive funds. Expenses for travel and project activities are used for the sake of promoting international research collaboration and enhancing global visibility. External grants obtained and used for research activities are listed in Appendix2-6.

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Appendix 1-5-2. FY2015 Project Expenditures (the exchange rate used: 1USD= JPY)

i) Overall project funding

Cost Items	Details	Costs (million yen)
Personnel	Center director and Administrative director	45
	Principal investigators (no. of persons = 12):	73
	Other researchers (no. of persons = 32):	194
	Research support staffs (no. of persons = 7):	26
	Administrative staffs (no. of persons = 14):	56
	Total	394
Project activities	Gratuities and honoraria paid to invited principal investigators (no. of persons):	0
	Cost of dispatching scientists (no. of persons = 3):	15
	Research startup cost (no. of persons = 8):	17
	Cost of satellite organizations (no. of satellite organizations = 2):	17
	Cost of international symposiums (no. of symposium = 1):	3
	Rental fees for facilities	54
	Cost of consumables	11
	Cost of utilities	63
	Other costs	20
	Total	200
Travel	Domestic travel costs	1
	Overseas travel costs	6
	Travel and accommodations cost for invited scientists (no. of domestic scientists): (no. of overseas scientists):	1
	Travel cost for scientists on secondment (no. of domestic scientists): (no. of overseas scientists):	0
	Total	8
Equipment	Depreciation of buildings	
	Depreciation of equipment	425
	Total	425
Other research projects	Projects supported by other government subsidies, etc.	
	Commissioned research projects, etc.	24
	Grants-in-Aid for Scientific Research, etc.	
	Total	24
Total		1051

million yen

WPI grant	519
Costs of establishing and maintaining facilities	0
Establishing new facilities (Number of facilities: , m²) Costs paid:	
Repairing facilities (Number of facilities: , m²) Costs paid:	
Others	
Cost of equipment procured	11
2 Draft chambers	4
1 Desktop fume hood	3
Installation of lab benches with chemical storage	2
Others	2

ii) Costs of Satellites and Partner institutions

Cost Items	Details	Costs (million yen)
Personnel	Principal investigators (no. of persons):	
	Other researchers (no. of persons): 3	
	Research support staffs (no. of persons):	
	Administrative staffs (no. of persons):	
	Total	17
Project activities		
Travel		
Equipment		
Other research projects		
Total		17

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Appendix 2-1. List of papers underscoring each research achievement

- List papers underscoring each research achievement listed in the item 2-1 "Research results to date" (up to 40 papers) and provide a description of the significance of each (within 10 lines).
- For each, write the author name(s); year of publication; journal name, volume, page(s), and article title. Any listing order may be used as long as format is the same.
- If a paper has many authors, underline those affiliated with the Center.
- If a paper has many authors (say, more than 10), all of their names do not need to be listed.

[2] Reciprocal interaction between the extended amygdala and arousal systems (Sakurai/ Sakaguchi Lab)

1. Sakurai T (2014) *Nat Rev Neurosci* **15**: 719-731. The role of orexin in motivated behaviours.

   An invited review about roles of orexin in regulation of motivated behaviors and reward-seeking behaviors, which summarizes mechanisms by which orexin neurons are activated in response to emotionally-salient cues, and how orexin system support motivated behavior.

2. Hasegawa E, Yanagisawa M, Sakurai T, Mieda M (2014) *J Clin Invest* **124**: 604-616. Orexin neurons suppress narcolepsy via 2 distinct efferent pathways.

   We showed that targeted restoration of orexin receptor expression by AAV-mediated gene transfer in noradrenergic neurons of the locus coeruleus (LC) and in serotonergic neurons of the dorsal raphe (DR) in Ox1r -/-;Ox2r-/- mice, which display a severe narcoleptic phenotype, differentially inhibited fragmentation of wakefulness and cataplexy, respectively, suggesting that DR serotonergic and LC noradrenergic neurons play differential roles in orexin neuron-dependent regulation of sleep/wakefulness.

3. Soya S, Shoji H, Hasegawa E, Hondo M, Miyakawa T, Yanagisawa M, Mieda M, Sakurai T (2013) *J Neurosci* **33**: 14549-14557. Orexin receptor-1 in the locus coeruleus plays an important role in cue-dependent fear memory consolidation.

   Ox1r-/- mice showed impaired freezing responses in both cued and contextual fear-conditioning paradigms. Locus coeruleus noradrenergic (LC-NA) neurons are activated after test sessions of both cued and contextual tests, but numbers of activated cells were significantly fewer in Ox1r-/- mice. AAV-mediated expression of OX1R in LC-NA neurons in Ox1r-/- mice restored the freezing behavior to the auditory cue to a comparable level to that in wild-type mice in the test session. These observations suggested that OX1R in LC-NA neurons plays an important role in cue-dependent fear memory formation and/or retrieval.

[3] The causal role of the mesolimbic brain system in the control of sleep and wakefulness (Lazarus Lab)

4. Lazarus M, Huang ZL, Lu J, Urade Y, Chen JF (2012) *Trends Neurosci* **35**: 723-732. How do the basal ganglia regulate sleep–wake behavior?

   In this paper we discussed anatomical and molecular mechanisms of sleep-wake regulation in the basal ganglia (BG), and proposed that adenosine and dopamine receptors in the nucleus accumbens (NAc) are involved in the integration of behavioral processes and the induction of wakefulness through cortical activation.

5. Lazarus M, Chen JF, Urade Y, Huang ZL (2013) *Curr Opin Neurobiol* **23**: 780-785. Role of the basal ganglia in the control of sleep and wakefulness

   The basal ganglia (BG) act as a cohesive functional unit that regulates motor function, habit formation, and reward/addictive behaviors, but the debate has only recently started on how the BG maintain

wakefulness and suppress sleep to achieve all these fundamental functions of the BG. Neurotoxic lesioning, pharmacological approaches, and the behavioral analyses of genetically modified animals revealed that the striatum and globus pallidus are important for the control of sleep and wakefulness. In both reviews (2. and 3.), the authors discuss anatomical and molecular mechanisms for sleep-wake regulation in the BG and propose a plausible model in which the nucleus accumbens integrates behavioral processes with wakefulness through adenosine and dopamine receptors.

6. Lazarus M, Shen HY, Cherasse Y, Qu WM, Huang ZL, ..., Hayaishi O, Urade Y, Chen JF (2011) *J Neurosci* **31**: 10067-10075. Arousal Effect of Caffeine Depends on Adenosine A2A Receptors in the Shell of the Nucleus Accumbens.

   Acting opposite to adenosine, caffeine enhances wakefulness. By using powerful tools for site-specific gene manipulations, such as conditional knockout mice for the adenosine A2A receptors (A2AR) based on the Cre/lox technology, or local infection with adeno-associated virus carrying short-hairpin RNA of A2AR to silence their expression, the specific role of A2AR in the basal ganglia was investigated. Deletion of A2AR selectively in the nucleus accumbens (NAc) shell was found to result in abrogation of caffeine's effect on wakefulness. Excitatory A2AR on neurons within the shell of the NAc must be tonically activated by adenosine for caffeine to be effective as an A2AR antagonist. This line of research indicates that activation of A2ARs on neurons in the NAc may contribute to the restraint of the arousal system, whereby caffeine can override the "adenosine brake" to promote wakefulness.

[4] Cortical neural networks in slow-wave sleep (Greene/Vogt Lab)

7. Bjorness T, Dale N, Mettlach G, Fienberg A, Sonneborn A, Sahin B, Yanagisawa M, Bibb H, Greene R (2016) *J Neurosci* **36**(13): 3709–3721. An Adenosine-Mediated Glial-Neuronal Circuit for Homeostatic Sleep. **J Neuroscience* featured publication

   In this paper we describe the role of adenosine signaling in regulating the buildup of sleep need during waking and the release of sleep pressure during slow wave activity. It shows that slow wave activity is a central element in sleep regulation. We aim to further investigate the mechanisms underlying slow wave activity and its regulation.

8. Jaafari N, Vogt KE, Saggau P, Leslie LM, Zecevic D, Canepari M. (2015) *Adv Exp Med Biol* **859**:103-25. Optical investigaion of neuronal circuits. Combining Membrane Potential Imaging with Other Optical Techniques.

   In this review we summarize our techniques to refine membrane potential imaging and to combine it with other optical techniques, such as release of caged pharmacological compounds and with optogenetics.

9. Willadt S, Canepari M, Yan P, Loew LM, Vogt KE. (2014) *Front Cell Neurosci* **8**:311. Combined optogenetics and voltage sensitive dye imaging at single cell resolution.

   This paper describes one of the first successful attempts to analyze a neural circuit with purely optical methods. By combining voltage imaging and optogenetics we can stimulate specific neurons and at the same time record the response of the network they are embedded in. We are planning to adapt these techniques to the in-vivo situation.

[5] Interaction of sleep, memory and adult neurogenesis (Sakurai/Sakaguchi Lab)

10. Sakaguchi M and Okano H. (2012) *Dev Neurobiol* **72**(7):1059-67. Neural stem cells, adult neurogenesis and galectins: from bench to bedside.

    We have been studying the clinical use of Galectin-1 for curing several neural diseases, for example, spinal cord injury and brain ischemia. We have found ameliorative effects of the protein in the model of those diseases including one in primate. We have discussed potential barriers when we try

to deploy the knowledge obtained in small mammals to human.

11. Hirota Y, Sawada M, Kida Y, Huang SH, Yamada O, Sakaguchi M, Ogura T, Okano H, Sawamoto K. (2012) *Stem Cells* **30**(8):1726-33. Roles of planar cell polarity signaling in maturation of neuronal precursor cells in the postnatal mouse olfactory bulb.

    Our group has been integrating knowledge and techniques in multi-disciplinary research fields, such as adult neurogenesis, sleep science and memory. Dr. Sakaguchi has been in collaboration with Dr. Sawamoto's and Dr. Okano's groups for long-term, which helps our group to gather current information in the adult neurogenesis field. In fact, our group is now collaborating with Dr. Okano's group to examine primate model of Parkinson's disease for its sleep symptoms. It will help our group to progress the finding in mouse model to pre-clinical stage in the future.

12. Sakaguchi M and Hayashi Y. (2012) *Mol Brain* **5**:32. Catching the engram: strategies to examine the memory trace

    Engram is one of the focus of memory research. Recent technical advances in genetics and optics have made significant progresses in finding the physical entity of memory - the so-called memory engram. The paper aimed at providing a comprehensive overview of the research to find the memory engram. Our group is now utilizing several advanced technique including optogenetics, which the paper laid out some ideas of use for sleep and memory research.

13. Arruda-Carvalho M, Sakaguchi M, Akers KG, Josselyn SA, Frankland PW. (2011) *J Neurosci* **31**(42):15113-27. Post-training ablation of adult-generated neurons degrades previously-acquired memories.

    In this paper, we provided an evidence that the adult-born neurons in the hippocampus could encode a memory. Selective removal of a population of predominantly mature, adult-generated neurons before learning did not affect the formation of new contextual fear or water maze memories, whereas removal of an equivalent population after learning degraded existing memories without affecting nonhippocampal memory. Ablation of these adult-generated neurons even 1 month after learning produced equivalent memory degradation.

14. Arruda-Carvalho M, Akers KG, Guskjolen AJ, Sakaguchi M, Josselyn S, Frankland PW. (2014) *J Neurosci* **34**(47):15793-803. Post-training ablation of adult-generated olfactory granule cells degrades odor-reward memories.

    In addition to the hippocampal adult-born neurons, this paper has shown that the adult-born neurons in the olfactory system could also encode memory information after learning. Since odor associated memory has been repeatedly used to reactivate memory during non-REM sleep, our group is now planning to implement the odor-evoked memory reactivation by stimulating adult-born neurons during sleep to clarify the mechanisms of memory consolidation during sleep.

15. Fujinaka A, Li R, Hayashi M, Kumar D, Changarathil G, Naito K, Miki K, Nishiyama T, Lazarus M, Sakurai T, Kee N, Nakajima S, Wang SH, Sakaguchi M. (2016) *Mol Brain* **9**:2. Effect of context exposure after fear learning on memory generalization in mice.

    We have discovered that there is a vulnerable time period in which a fearful memory could be associated with other non-harmful memory. It suggests that when people experienced tremendous amount of psychological stress, there would be a need to care those people to avoid exposing to risk factors which could be generalized to the traumatic memory. Recently, out group found an interesting correlation of this phenomena with sleep.

16. Sakaguchi M, Kim K, Yu LMY, Hashikawa Y, Sekine Y, Okumura Y, Kawano M, Hayashi M, Kumar D, Boyden ES, McHugh TJ, Hayashi Y. (2015) *PLoS ONE* **10**(6): e0130163. Inhibiting the activity of CA1 hippocampal neurons prevents the recall of contextual fear memory in inducible ArchT transgenic mice.

We have developed a new transgenic mouse line, in which a light sensitive protein, ArchT, can be expressed by a convenient and popular genetic engineering switch, tTA. Using this elegant technique, the paper also showed that memory retrieval could be transiently suppressed by shining light to the neurons in the hippocampus where the ArchT expressing neurons participated in memory. The mice are currently employed to transiently inhibit reactivation of memory during sleep. By the doing so, we are aiming at providing a direct evidence of functional correlation between memory reactivation and sleep.

[6] Elucidation of the function of REM sleep (Hayashi Lab)

17. Hayashi Y (co-corresponding author), Kashiwagi M, Yasuda K, Ando R, Kanuka M, Sakai K, Itohara S (co-corresponding author). (2015) *Science* **350**: 957-961. Cells of a common developmental origin regulate REM/non-REM sleep and wakefulness in mice.

    In this paper, we identified a brain region important for switching from REM sleep to NREM sleep, and established transgenic mice where REM sleep can be artificially suppressed. Using this novel mouse model, we showed that REM sleep promotes slow wave activity during non-REM sleep. Slow waves are known to promote memory consolidation and synaptic plasticity, and thus our study implicates that REM sleep is important for these events.

[9] Design and synthesis of orexin agonists (Nagase and Yanagisawa/Funato Lab)

18. Saitoh T, Nagase H. (2016) *MEDCHEM NEWS* **26**:90–96. Design and Synthesis of Non-peptidic Orexin Receptor Agonists.

    This paper is a review for the Award for Excellent Presentation in 33rd Medicinal Chemistry Symposium, DMC, PSJ in 2015. This paper described that the detailed process of discovery of **YNT-185, first orexin 2 receptor agonist** which has been reported in *J. Med. Chem.* paper and its pharmacological effects; *in vitro* and *in vivo* in wild-type mice.

19. Nagahara T, Saitoh T, Kutsumura N, Irukayama-Tomobe Y, Ogawa Y, Kuroda D, Gouda H, Kumagai H, Fujii H, Yanagisawa M, Nagase H. (2015) *J Med Chem* **58**:7931–7937. Design and Synthesis of Non-Peptide, Selective Orexin Receptor 2 Agonists. *Selected as a "Featured Article"

    Orexins are a family of neuropeptides that regulate sleep/wakefulness, acting on two G-protein-coupled receptors, orexin receptors 1 (OX1R) and 2 (OX2R). Genetic and pharmacologic evidence suggested that orexin receptor agonists, especially OX2R agonist, will be useful for definitive remedy of the sleep disorder narcolepsy/cataplexy. Our paper reported the discovery of a potent (EC_{50} on OX2R is 23 nM) and OX2R-selective (OX1R/OX2R EC_{50} ratio is 70) agonist, 4'-methoxy-*N*,*N*-dimethyl-3'-[*N*-(3-{[2-(3-methylbenzamido)ethyl]amino}phenyl)sulfamoyl]-(1,1'-biphenyl)-3-carboxamide (**YN-1055**, the compound **26** in this paper). In addition, this paper also reported the water-soluble OX2R agonist, **YNT-185** dihydrochloride (the compound **31** in this paper) was developed. **YNT-185** showed the potent agonistic activity (EC_{50} on OX2R is 28 nM) as same as that of **YN-1055**, and the *in vivo* assay of its dihydrochloride **31** showed definite wake-promoting effects.

[10] Identification of somnogenic natural compounds and elucidation of their mechanisms (Urade Lab)

20. Chen CR, Zhou XZ, Luo YJ, Huang ZL, Urade Y, Qu WM. (2012) *Neuropharmacology* **63**: 1191-1199. Magnolol, a major bioactive constituent of the bark of Magnolia officinalis, induces sleep via the benzodiazepine site of GABA(A) receptor in mice.

    Magnolol, an active ingredient of the bark of *Magnolia officinalis*, has been reported to exert potent anti-epileptic effects via the $GABA_A$ receptor. Magnolol significantly shorten the sleep latency, increase the amount of non-rapid eye movement (non-REM, NREM) and rapid eye movement (REM) sleep for

3 h after administration with an increase in the number of NREM and REM sleep episodes. Immunohistochemical study showed that magnolol increased c-Fos expression in the neurons of ventrolateral preoptic area, a sleep center in the anterior hypothalamus, and decreased c-Fos expression in the arousal tubero-mammillary nucleus, which was located in the caudolateral hypothalamus.

21. Liu Z, Xu XH, Liu TY, Hong ZY, Urade Y, Huang ZL, Qu WM. (2012) *CNS Neurosci Ther* **18**: 623-630. Safranal enhances non-rapid eye movement sleep in pentobarbital-treated mice.

    Safranal is an active ingredient in the saffron, which is used in traditional medicine. It has been reported to have sedative and anti-epileptic effects. Safranal increased the duration of non-rapid eye movement sleep (NREM), shortened NREM sleep latency, and enhanced the delta power activity of NREM sleep in the presence of a low dose of pentobarbital 20 mg/kg. Immunohistochemical evaluation revealed that safranal increased c-Fos expression in the ventrolateral preoptic nucleus (VLPO), one of the putative sleep centers, and decreased it in the arousal histaminergic tuberomammillary nuclei (TMN).

22. Masaki M, Aritake K, Tanaka H, Shoyama Y, Huang ZL, Urade Y. (2012) *Mol Nutr Food Res* **56**: 304-308. Crocin promotes non-rapid eye movement sleep in mice.

    Crocus sativus L. (saffron) has been traditionally used for the treatment of diseases in the nervous systems. We found crocin, a major natural form of saffron, increased the total time of non-rapid eye movement sleep at doses of 30 and 100 mg/kg, without change the amount of rapid eye movement sleep nor show any adverse effects, such as rebound insomnia, after the induction of sleep.

23. Omori K, Kagami Y, Yokoyama C, Moriyama T, Matsumoto N, Masaki M, Nakamura H, Kamasaka H, Shiraishi K, Kometani T, Kuriki T, Huang ZL, Urade Y. (2012) *Sleep Biol Rhythms* **10**: 38-45. Promotion of non-rapid eye movement sleep in mice after oral administration of ornithine.

    Ornithine, a free amino acid that is not genetically coded to be used for protein synthesis, is used to attenuate physiological fatigue. We examined the sleep promoting effects of ornithine and found ornithine (1.0 and 3.0 g/kg of body weight) increased the amount of non–rapid eye movement sleep without affecting the power spectrum density of non–rapid eye movement sleep.

24. Qu WM, Yue XF, Sun Y, Fan K, Chen CR, Hou YP, Urade Y, Huang ZL. (2012) *Br J Pharmacol* **167**: 587-598. Honokiol promotes non-rapid eye movement sleep via the benzodiazepine site of the GABA(A) receptor in mice.

    Honokiol, an active component of the Chinese herb houpo, is used to treat depression effectively in China. Honokiol (10 and 20 mg/kg) significantly shortened the sleep latency to non-rapid eye movement (non-REM, NREM) sleep and increased the amount of NREM sleep. Honokiol increased the number of state transitions from wakefulness to NREM sleep and, subsequently, from NREM sleep to wakefulness. Honokiol increased c-Fos expression in ventrolateral preoptic area (VLPO) neurons, as examined by immunostaining, and excited sleep-promoting neurons in the VLPO by whole-cell patch clamping in the brain slice.

25. Cho S, Yoon M, Pae AN, Jin YH, Cho NC, Takata Y, Urade Y, Kim S, Kim JS... Huang ZL. (2014) *Psychopharmacology (Berl)* **231**: 2825-2837. Marine polyphenol phlorotannins promote non-rapid eye movement sleep in mice via the benzodiazepine site of the $GABA_A$ receptor.

    Polyphenol compounds of herbal medicines has been considered as an important sedative–hypnotic compound. Marine plants have not been recognized as a potential source of novel, although they have marine poly-phenols, known as phlorotannins. Phlorotannin preparation significant decrease in sleep latency and an increase in the amount of non-rapid eye movement sleep measured by recording electroencephalograms (EEG) and electromyograms. The hypnotic effects of PRT were completely abolished by pretreatment with flumazenil.

26. Cherasse Y, Saito H, Nagata N, Aritake K, Lazarus M, Urade Y. (2015) *Mol Nutr Food Res* **59**: 2087-

2093. Zinc-containing yeast extract promotes nonrapid eye movement sleep in mice.

Zinc is an essential trace element for humans and animals, being located, among other places, in the synaptic vesicles of cortical glutamatergic neurons and hippocampal mossy fibers in the brain. Extracellular zinc has the potential to interact with and modulate many different synaptic targets, including glutamate and GABA receptors. Because of the central role of these neurotransmitters in brain activity, we examined in this study the sleep-promoting activity of zinc by monitoring locomotor activity and electroencephalogram after its administration to mice. Zinc-containing yeast extract (40 and 80 mg/kg) dose dependently increased the total amount of nonrapid eye movement sleep and decreased the locomotor activity.

27. Monoi N, Matsuno A, Nagamori Y, Kimura E, Nakamura Y, Oka K, Sano T, Midorikawa T, Sugafuji T... Urade Y. (2016) *J Sleep Res* **25**: 116-123. Japanese sake yeast supplementation improves the quality of sleep: a double-blind randomised controlled clinical trial.

Adenosine promotes sleep/wake regulation via A1 and A2a receptors. Japanese sake yeast, but not beer yeast and baker's yeast, is interestingly enriched in adenosine analogues activates A2a receptors in *vitro*. In a double-blind placebo-controlled crossover clinical study, electroencephalogram analyses revealed that sake yeast supplementation significantly increased delta power during the first cycle of slow-wave sleep. Sake yeast supplementation also significantly increased growth hormone secretion in the urine on awakening. Subjective sleepiness and fatigue in the morning were improved by sake yeast.

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Appendix 2-3. List of the cooperative research agreements in and outside Japan

1. Counterpart of an Agreement : Akita University
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : April, 2013
   Summary of an Agreement : We have concluded a service agreement with Shimizu, a clinician at the Akita University Graduate School of Medicine Doctoral Course in Medicine/Bioregulatory Medicine Department of Neuropsychiatry and engaged in joint research in the pathological study of sleep disorders, translational research, etc. Concerning narcolepsy, Akita University is the sole facility capable of determining orexin concentrations in cerebrospinal fluid and accumulating samples, as well as clinical information on various cases. Furthermore, to facilitate human molecular genetic research into short sleepers, short-sleeper candidates were sought within the university to confirm short-term sleep of six candidates by analysis with ActiGraph. Two cases are expected as family and blood collection, linkage analysis, exome analysis, etc. will be performed with the one case for which consent was obtained.

2. Counterpart of an Agreement : University of Texas Southwestern Medical Center (UTSW)
   Name of an Agreement : Collaboration Research Agreement and Sponsored Research agreement
   Dates of an Agreement : November, 2013
   Summary of an Agreement : The UTSW has been the research center of Institute Director, Masashi Yanagisawa, for more than 20 years and has nurtured a close relationship as an IIIS satellite. The four satellite PIs (J. Takahashi, R. Greene, C. Green and Q. Liu) and joint research with the respective satellite PIs has been conducted since establishing IIIS, as well as concluding joint research or sponsored research agreements when research funds were provided. Liu engaged in research into intracellular signal transduction of sleep control nerve cells by analyzing phosphoprotein using mass spectrometric techniques and joint research into the molecular control mechanism of essential terror by forward genetics using an ENU mutant mouse, C. Green engaged in sponsored research into RNA analysis of sleep-deprived mice, R. Greene covered sponsored research on sleep homeostasis and the sleep-awakening control of adenosine, while Takahashi is currently undertaking joint research on rhythm control of sleep. To further enhance collaboration with Takahashi, we plan to promote personnel exchange by jointly applying for the ”JSPS Program for Advancing Strategic International Networks to Accelerate the Circulation of Talented Researchers” next year. Institute Director, Yanagisawa, concurrently serves as Professor at the University of Tsukuba and UTSW as a joint appointment (95:5) between both universities and intellectual property rights belonging to the Institute Director go to both universities based on this joint appointment.

3. Counterpart of an Agreement : RIKEN Brain Science Institute
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : April, 2014
   Summary of an Agreement : From 2013 on an ongoing basis, we have engaged in joint research with Dr. Shigeyoshi Itohara of the RIKEN Brain Science Institute, Laboratory for Behavioral Genetics, to elucidate the physiological significance of sleep toward developing therapy for mental disorders.

4. Counterpart of an Agreement : Tokyo Medical University
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : April, 2014
   Summary of an Agreement : It had been known that one of the molecular targets of thalidomide related to teratogenic effect is cereblon. We investigate whether the hypnosis action goes through the same molecules target.

5. Counterpart of an Agreement : RIKEN Bioresource Center
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : September, 2014

Summary of an Agreement : Following on from 2014, we have engaged in joint research into "genetic mapping analysis of mutant mouse obtained in ENU screen" this year, the results of which help identify candidate genes with potential to access the molecular control mechanism of essential terror. We plan to maintain this relationship in future as well as continuing focused collaboration as research progresses.

6. Counterpart of an Agreement : Marine Ecology Research Institute
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : December, 2014
   Summary of an Agreement : We perform the development of the sleep measurement device monitoring the electroencephalograph in fish.

7. Counterpart of an Agreement : National Institute of Advanced Industrial Science and Technology
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : April, 2015
   Summary of an Agreement : Molecular studies on the effect of *Ashwagandha* extracts on sleep quality and rhythm

8. Counterpart of an Agreement : Nagaoka University of Technology
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : April, 2015
   Summary of an Agreement : We have engaged in joint research into the "application and verification of brain-wave fractal analysis to sleep diagnosis" with Dr. Masahiro Nakagawa of the Graduate School of Nagaoka University of Technology. The purpose of this research is to optimize the brain-wave fractal analysis developed by the Nagaoka University of Technology to apply to the analysis and diagnosis of sleep and establish a new means of sleep diagnosis by developing a device to measure brain waves which does not disturb sleep. We are jointly attempting to acquire public research funds.

9. Counterpart of an Agreement : Graduate School of Pharmaceutical Science, Kyoto University
   Name of an Agreement : Collaboration Research Agreement
   Dates of an Agreement : July, 2015
   Summary of an Agreement : We have installed Hitoshi Okamura of the Department of System Biology, Kyoto University Graduate School of Pharmaceutical Sciences as a satellite PI and concluded a joint research agreement to commence joint research into sleep and biological rhythms. The purpose of this joint research is to seek out the time-difference controlling gene by ENU random mutant screening, and we are currently preparing to install rearing equipment (rack for circadian rhythm measurement) in the animal facility.

10. Counterpart of an Agreement : Center for Genomic Medicine, Kyoto University
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : December, 2015
    Summary of an Agreement : We have concluded a joint research agreement with Dr. Fumihiko Matsuda of the Center for Genomic Medicine, Kyoto University as part of moves to commence joint research into genome epidemiologic study of genetic/environmental factors related to human sleep. The purpose of this research is to analyze the samples and information of the "Nagahama Cohort Study", and seek for human counterparts of the sleep-related genes obtained from mice in IIIS. Also, conversely, if the dyssomnia gene is identified in the "Nagahama Cohort Study," the responsible mutations will be verified with a mouse. This is a joint research with efficient mediation between basic and clinical results to ensure efficient progress. We invited Dr. Moho Morita, who has various achievements in a genome cohort study, to IIIS as a visiting associate professor.

11. Counterpart of an Agreement : Hoshi University
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : November, 2015
    Summary of an Agreement : We will perform molecular and pharmacological analyses of the interaction between the orexin receptors and the opioid receptors.

12. Counterpart of an Agreement : Fujifilm Corporation
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : January, 2014
    Summary of an Agreement : From 2013 to 2015, within the framework of ”excellent sleep consortium” as a consignment study of the Ministry of Agriculture, Forestry and Fisheries, Urade engaged in a joint research into ”pharmacokinetics of sleep improver and sleep mechanism elucidation” with Fujifilm and found the sleep enhancement action of zinc. The results of the joint research by Urade fruited in the market in the form of the sleep enhancement supplement “Suttone.” From 2016, a joint research by Nagase and Fujifilm will start.

13. Counterpart of an Agreement : Watanabe Oyster Institute
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : February, 2014
    Summary of an Agreement : The effect of new antioxidant material, discovered from oyster in the, on sleep has been investigated using a mouse model.

14. Counterpart of an Agreement : Mikasa Seiyaku Co.Ltd.
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : April, 2014
    Summary of an Agreement : We have engaged in the ongoing joint research with Mikasa Seiyaku on “external preparations containing nalfurafine.” This research targets the life-cycle management of nalfurafine (Nagase is one of the inventors), which is placed on the market by examining preparation methods. Two pharmaceutical patents have already been filed and with the intention to return the research results to society, we engage in licensing negotiations with some pharmaceutical companies.

15. Counterpart of an Agreement : Toray Industries, Inc.
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : May, 2014
    Summary of an Agreement : We have continued “research into sleep of nalfurafine hydrochloride and analog compound” with Toray since 2013. The pharmacology team of Yanagisawa Lab performs in-vitro and in-vivo evaluations of drug efficacy of nalfurafine hydrochloride on sleep, and an analog compound is synthesized by Nagase Lab jointly with Toray. Following this joint research, two patents have been already filed.

16. Counterpart of an Agreement : Lion Corporation
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : May, 2014
    Summary of an Agreement : We have engaged in ongoing joint research with Lion to “elucidate the mechanism of action of sleep quality-improving material.” This research resulted in the market as the sleep-improving supplement “Gussumin.”

17. Counterpart of an Agreement : Nisshin Seifun Group
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : Autumn, 2014
    Summary of an Agreement : We will investigate the food material related to the wheat with activity on sleep using a mouse model.

18. Counterpart of an Agreement : Hospital Department of Ibaraki Prefecture
    Name of an Agreement : Special Collaborative Research Agreement
    Dates of an Agreement : March, 2015
    Summary of an Agreement : Over five years from 2015, we conducted a joint project with the Hospital Department of Ibaraki Prefecture intended to “promote clinical research into sleep disorders” (sleep apnea syndrome in particular). Satoh Lab was installed in IIIS and we have engaged in research for sleep apnea and other sleep disorders. In future, the Satoh Lab will be expanded into a collaborative research department with Seven Dreamers Laboratories Inc.

19. Counterpart of an Agreement : Nishikawa Sangyo Co. Ltd.
    Name of an Agreement : Collaborative Research Agreement
    Dates of an Agreement : April, 2015
    Summary of an Agreement : We continue the joint research with Nishikawa Sangyo on "the impact of bedding on sleep" in 2016. Although experience on a daily basis reaffirms the significant impact of bedding on sleep, given the lack of scientific research and verification, we aim to objectively evaluate the influence of bedding on sleep. A correct and detailed evaluation of the influence of bedding currently centers on using bedding with varying ability to disperse body pressure.

20. Counterpart of an Agreement : Sumitomo Dainippon Pharma. Co. Ltd.
    Name of an Agreement : Collaborative Research Agreement
    Dates of an Agreement : March, 2016
    Summary of an Agreement : We commenced "exploratory research into remedies using a model mouse with REM sleep behavior disorder (RBD)" from 2016 with Sumitomo Dainippon Pharma. On March 31, 2016, we concluded a joint research agreement. Normally, in REM sleep, the tonus of the skeletal muscles of the whole body is controlled but RBD is a sleep disorder resulting in a lack of control of muscle tone due to some cause, whereupon the body starts moving while in REM sleep. Potential underlying causes may include Parkinson's disease, Lewy body disease, etc., although the cause of this disease is unknown. We seek for therapeutic agents using RBD model mice uniquely reared at IIIS and examine existing agents and chemical compounds provided by Sumitomo Dainippon Pharma.

21. Counterpart of an Agreement : JAXA Space Biomedical Research Office
    Name of an Agreement : Collaboration Research Agreement
    Dates of an Agreement : March, 2016
    Summary of an Agreement : The Promotion of Science Grants-in-Aid for Scientific Research for research into an innovative area, "Integral Understanding of Life-regulation mechanism from Space," which was jointly applied for by 11 groups, featuring Astronaut Furukawa as representative, was adopted and since August 2015, we have started joint research with the JAXA Space Biomedical Research Office. In 2015, an experiment of placing two-week closed environmental loads to narrow down stress marker candidates effectively reflected stress conditions in a closed environment, with adaptation training equipment installed in the astronauts' training building of JAXA. IIIS obtained ActiGraph and EEG data, and analyzed the sleep medical questionnaire to examine the impact on sleep of stress from closure.

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Appendix 2-4. Major Awards, Invited Lectures, Plenary Addresses (etc.)

1. Major Awards

- List main internationally-acclaimed awards received/unofficially announced in order from the most recent.
- For each, write the recipient's name, name of award, and year issued.
  In case of multiple recipients, underline those affiliated with the center.

1) Masashi Yanagisawa, The Walter B. Cannon Memorial Award (American Physiological Society), 2015

2) Akiyoshi Fukamizu, 19th Jokichi Takamine Memorial Award (The Society of Cardiovascular Endocrinology and Metabolism), 2015

3) Hiroshi Nagase, Yamazaki Teiichi Prize (Foundation for Promotion of Material Science and Technology of Japan), 2014

4) Yoshihiro Urade, Award for Distinguished Contributions (Chinese Sleep Research Societ), 2014

5) Masashi Yanagisawa, 17th JokichiTakamine Memorial Award (The Society of Cardiovascular Endocrinology and Metabolism), 2013

6) Hiroshi Nagase, National Commendation for Invention – the Invention Prize (Japan Institute of Invention and Innovation), 2013

7) Hiroshi Nagase, Okochi Memorial Technology Prize (Okochi Memorial Foundation), 2013

8) Junichi Hayashi, 24th Tsukuba Award (The Science and Technology Promotion Foundation of Ibaraki), 2013

9) Joseph Takahashi, Outstanding Scientific Achievement Award (Sleep Research Society), 2012

10) Takeshi Sakurai, The Commendation for Science and Technology by the Minister of Education, Culture, Sports, Science and Technology (Ministry of Education, Culture, Sports, Science and Technology), 2012

2. Invited Lectures, Plenary Addresses (etc.) at International Conferences and International Research Meetings

- List up to 10 main presentations in order from most recent.
- For each, write the lecturer/presenter's name, presentation title, conference name and date(s)

1) Michael Lazarus, "A nucleus accumbens circuit in control of sleep", The 4th International Pediatric Sleep Association Congress (Taipei, Taiwan), 2016

2) Yu Hayashi, "Identification of a sleep regulatory circuit and implications for the function and evolution of REM sleep", JST CREST-PRESTO Joint International Symposium (Tokyo, Japan), 2015

3) Masashi Yanagisawa, Plenary Session "Toward solving the mystery of sleep: From reverse genetics to forward genetics in mice", Sleep2015 (Seattle, USA), 2015

4) Hiroshi Nagase, "Design and synthesis of novel δ opioid agonists and their pharmacologies", Drug Discovery & Therapy World Congress 2015 (Boston, USA), 2015

5) Joseph Takahashi, "Molecular Architecture of the Circadian Clock in Mammals", Chronobiology Symposium "Translation of circadian biology: Implications for the clinic" (Pennsylvania, USA), 2015

6) Yoshihiro Urade, "Why coffee wakes us up - Role of adenosine A2A receptors in the nucleus accumbens for sleep-wake regulation", World Congress on Sleep Medicine (Seoul, Korea), 2015

7) Yoshihiro Urade, "Key Roles of prostaglandin D2 and adenosine in sleep regulation: from pharmacological approaches to gene-knockout mice", The Joint Congress of the Inauguration Conference of the Asia Sleep Society of Medicine (ASSM) and The 8th National Congress of the Chinese Sleep Research Society (CSRS)(Beijing, China), 2014

8) Masashi Yanagisawa, "Forward genetics of sleep in ENU-mutagenized mice", Gordon Research Conference on Sleep Regulation & Function (Galveston, USA), 2014

9) Masashi Yanagisawa, "Forward genetics of sleep in mice", Recent Advances and Controversies in Measuring Energy Metabolism (Tokyo, Japan), 2014

10) Hiroshi Nagase, "Synthesis of a novel opioid receptor agonist, SYK-146 with 1,3,5-trioxazatriquinane skeleton and its pharmacologies", DPhG Jahrestagung 2014 (Frankfurt, Germany), 2014

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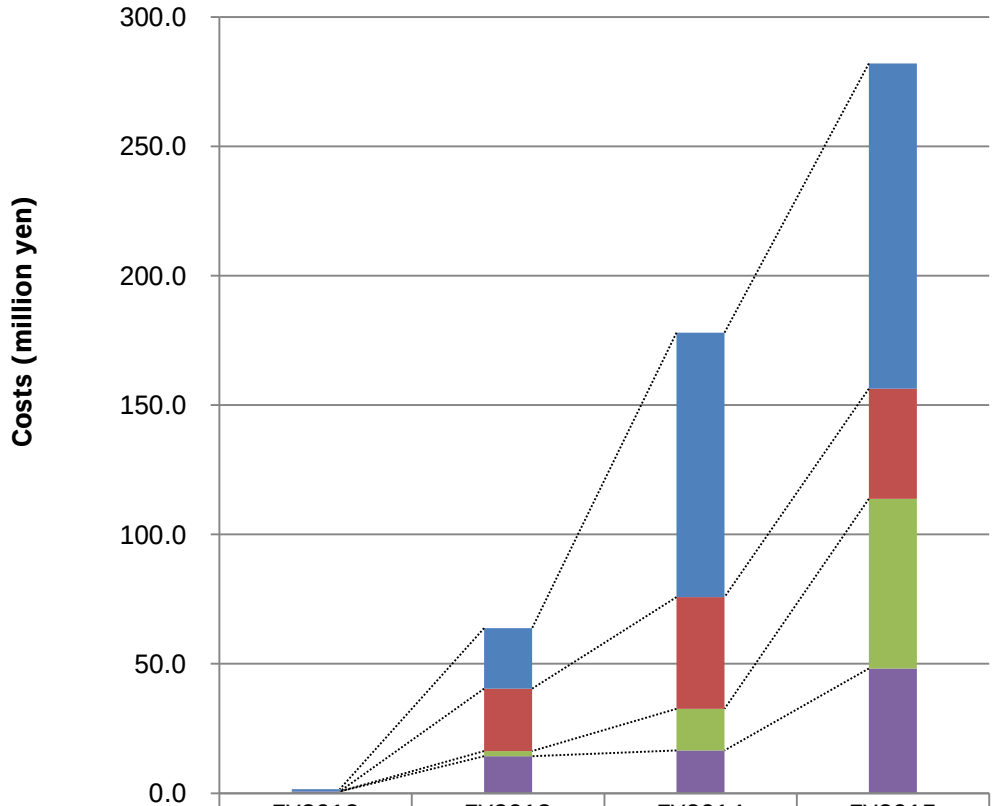
Appendix 2-6. Amounts of Non-WPI project funding (grants)

*Make a graph of the annual transition in non-WPI project funding (grants).

Annual Transition in the Amounts of Project Funding



	FY2012	FY2013	FY2014	FY2015
Grants-in-Aid for Scientific Research, etc.	0.9	23.5	102.2	125.8
Commissioned research projects, etc.	0.0	24.0	43.1	42.5
Joint researches, etc.	0.0	2.0	16.1	65.6
Grants and Endowments	0.7	14.3	16.5	48.2

- Graph doesn't include the FIRST program and university internal programs

- Describe external funding warranting special mention. Include the name and total amount of each grant.

FY2012

・ Japan Society for the Promotion of Science (JSPS) Funding Program for World-Leading Innovative R&D on Science and Technology (FIRST)
Total: ¥451,920,000 (Masashi Yanagisawa)

FY2013

・ Japan Society for the Promotion of Science (JSPS) Funding Program for World-Leading Innovative R&D on Science and Technology (FIRST)
Total: ¥397,624,215 (Masashi Yanagisawa)

・ Japan Science and Technology Agency (JST) Precursory Research for Embryonic Science and Technology (PREST)
Total: ¥15,600,000 (Yu Hayashi)

FY2014

- Japan Society for the Promotion of Science (JSPS) Grants-in-Aid for Scientific Research (S)
  Total: ¥43,940,000 (Masashi Yanagisawa)

- Japan Science and Technology Agency (JST) Precursory Research for Embryonic Science and Technology (PREST)
  Total: ¥14,300,000 (Yu Hayashi)

FY2015

- Japan Society for the Promotion of Science (JSPS) Grants-in-Aid for Scientific Research (S)
  Total: ¥39,000,000 (Masashi Yanagisawa)

- Japan Science and Technology Agency (JST) Precursory Research for Embryonic Science and Technology (PREST)
  Total: ¥9,360,000 (Yu Hayashi)

- Japan Society for the Promotion of Science (JSPS) Grants-in-Aid for Scientific Research on Innovative Areas
  Total: ¥28,470,000 (Hiroshi Nagase)

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Appendix 2-8. FY 2015 List of Project's Media Coverage

- Select main items of coverage, and list them within these 2 pages.

No.	Date	Type media (e.g., newspaper, television)	Description
1	May 2015	**Magazine** Medical Asahi, May 2015 issue	Series: Drugs born in Japan volume 10 "Bozentan" (Yanagisawa)
2	June 2015	**Magazine** Medical Asahi, June 2015 issue	Series: Drugs born in Japan volume 11 "Suvorexant" (Yanagisawa)
3	July 1, 2015	**Web** NASA Official Website	ISS Benefits For Humanity: Hope Crystallizes (Urade)
4	July 21, 2015	**Web** MyNavi News	A scientist of sleep research center in Tsukuba explained the mechanism of dreams depicted in the movie "Inside Out" (Yanagisawa)
5	Aug 5, 2015	**Web** nippon.com	Solving the mystery of sleep: A cutting-edge research center in Tsukuba led by a discoverer of orexin (Yanagisawa)
6	Aug 10, 2015	**Radio** Radio Nikkei 1, "Byoyaku Hour"	Regulation of sleep/wakefulness; orexin system as a drug target (radio lecture by Yanagisawa)
7	Aug 13, 2015	**Newspaper** The Mainichi	Towards the mystery of sleep: challenges of IIIS (Yanagisawa)
8	Sep 8, 2015	**Television** NHK World	"Medical Frontiers" Volume 5, Sleep (Yanagisawa)
9	Sep 24, 2015	**Newspaper** Nikkei Sangyo Shimbun	Front-line report of medical cares: Ibaraki Prefectural Medical Center of Psychiatry for sleep disorders (Satoh)
10	Sep 29 - Oct 1, 2015	**Newspaper** The Daily Engineering & Consruction News, The Yomiuri Shimbun, Web Medical Tribune Aging Style	International Institute for Integrative Sleep Medicine, University of Tsukuba: Construction of IIIS Building completed and the inauguration ceremony was held
11	Oct 1, 2015	**Newspaper** The Mainichi	Q & A: How to maintain good quality of sleep in the refuge (Yanagisawa answered an interview by newspaper)

No.	Date	Type media (e.g., newspaper, television)	Description
12	Oct 11, 2015	**Television** NHK	Science ZERO "Mystery of Sleep: The frontier of sleep medicine" (Yanagisawa)
13	Oct 22, 2015	**Web** Medical Daily	Our Dream State, REM Sleep, Influences Memory Consolidation During Other Sleep Phases (Hayashi)
14	Oct .22 – 27, 2015	**Newspaper / web** The Asahi Shimbun, The Yomiuri Shimbun, The Mainichi, The Nikkei Biglobe News, MyNavi News *And other 22 coverages	New insights into the role and function of REM sleep (Hayashi)
15	Oct 23, 2015	**Television** NHK News	New insights into the role and function of REM sleep (Hayashi, interview by telephone)
16	Dec 3, 2015	**Newspaper** The Asahi Shimbun	Discovery of a novel compound regulating wakefulness (Nagase)
17	Dec 5, 2015	**Newspaper** The Mainichi	Q: Does counting sheep help us sleeping? (Yanagisawa interview)
18	Dec 27, 2015	**Television** TBS	Yume-no-tobira plus "Dream compounds making us sleep well" (Urade)
19	Jan 8, 2016	**Magazine** Bungei Shunju Feb issue	Feature article: Qualification for the leaders making breakthroughs (Yanagisawa interview)
20	Jan 25 - Feb 4, 2016	**Newspaper** The Asahi Shimbun, The Joyo Shimbun **Web** Aging Style	Timing is important for the care of PTSD (Sakaguchi)
21	Feb 4, 2016	**Newspaper** The Yomiuri Shimbun	Feature article: orexin and sleep (Yanagisawa)
22	Feb 26, 2016	**Television** BS Fuji	Kakushin-no-ism (Yanagisawa)
23	Mar 14, 2016	**Newspaper** Nikkei Sangyo Shimbun	Innovative studies on substances regulating sleep (Yanagisawa)

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Appendix 3. List of papers of representative of interdisciplinary research activities

- List up to 10 papers that underscoring each interdisciplinary research activity and give brief accounts (within 10 lines).
- For each, write the author name(s); year of publication; journal name, volume, page(s), and article title. Any listing order may be used as long as format is the same.
  If a paper has many authors, underline those affiliated with the Center.
- If a paper has many authors (say, more than 10), all of their names do not need to be listed.

1. Lee IT, Chang AS, Manandhar M, Shan YL, Fan JM, Izumo M, Ikeda Y, Motoike T, Dixon S, Seinfeld JE, Takahashi JS, Yanagisawa M. (2015) *Neuron* **85**(5): 1086-1102. Neuromedin S-Producing Neurons Act as Essential Pacemakers in the Suprachiasmatic Nucleus to Couple Clock Neurons and Dictate Circadian Rhythms.

   In this study, authors showed that a subset of the suprachiasmatic nucleus (SCN) neurons expressing the neuropeptide neuromedin S (NMS) plays an essential role in the generation of daily rhythms in behavior.

2. Nagahara T, Saitoh T, Kutsumura N, Irukayama-Tomobe Y, Ogawa Y, Kuroda D, Gouda H, Kumagai H, Fujii H, Yanagisawa M, Nagase H. (2015) *J. Med. Chem*. **58**(20): 7931-7937. Design and Synthesis of Non-Peptide, Selective Orexin Receptor 2 Agonists.

   Genetic and pharmacologic evidence suggested that orexin receptor agonists, especially OX2R agonist, will be useful for definitive remedy of the sleep disorder narcolepsy/cataplexy. This paper reported the discovery of a potent and the orexin receptor (OX2R)-selective agonist which showed definite wake-promoting effects.

3. Cherasse Y, Saito H, Nagata N, Aritake K, Lazarus M, Urade Y. (2015) *Mol. Nutr. Food Res*. **59**(10): 2087-2093. Zinc-containing yeast extract promotes nonrapid eye movement sleep in mice.

   This paper showed that zinc, an essential trace element for humans and animals, can induce sleep. Zinc-containing yeast extract increased the total amount of NREM sleep and decreased the locomotor activity, whereas this preparation did not affect the amount of REM sleep or show rebound of insomnia. This study opened the way to new types of food supplements designed to improve sleep.

4. Wang ZQ, Liu SM, Kakizaki M, Hirose Y, Ishikawa Y, Funato H, Yanagisawa M, Yu YH, Liu QH. (2014) *J. Biol. Chem*. **289**(46): 31950-35959. Orexin/Hypocretin Activates mTOR Complex 1 (mTORC1) via an Erk/Akt-independent and Calcium-stimulated Lysosome v-ATPase Pathway.

   The lack of the neuropeptide orexin results in narcolepsy, but the downstream pathways of orexin signaling are not known. The authors showed that orexin activates the mTOR pathway, a central regulator of cell growth and metabolism, suggesting that the mTORC1 pathway functions downstream of orexin/GPCR signaling, which plays a crucial role in many physiological and metabolic processes.

5. Kardon AP, Plgar E, Hachisuka J, Snyder LM, Cameron D, Savage S, Cai X, Karnup S... Nagase H,... Ross SE et al. (2014) *Neuron* **82**: 573-586. Dynorphin Acts as a Neuromodulator to Inhibit Itch in the Dorsal Horn of the Spinal Cord.

   The neural basis for relieving itch by menthol and other counterstimuli is unclear, and the underlying neuromodulatory mechanisms are unknown. We characterized a specific population of spinal inhibitory interneurons (B5-I neurons) and showed that dynorphin, which is released from B5-I neurons, is a key neuromodulator of pruritus.

6. Kaneko K, Mizushige T, Miyazaki Y, Lazarus M, Urade Y, Yoshikawa M, Kanamoto R, Chinata K. (2014) *Am. J. Physiol. Regul. Integr. Comp. Physiol.* **306**(4): R265-R272. δ-Opioid receptor activation stimulates normal diet intake but conversely suppresses high-fat diet intake in mice.

   The central opioid system is involved in a broadly distributed neural network that regulates food intake. We showed that activation of central δ-opioid receptor not only stimulated normal diet intake

but conversely suppressed high-fat diet intake as well.

7. Xu Q, Xu XH, Qu WM, Lazarus M, Urade Y, Huang ZL. (2014) *Pharmacol Biochem Behav.* **116**: 129-136. A mouse model mimicking human first night effect for the evaluation of hypnotics.

   A first night effect (FNE) in human is characterized by increased sleep latency and decreased total sleep time in an unfamiliar environment, but the mechanism and treatment for this acute insomnia are unclear. We developed an FNE model by inducing acute insomnia in mice that have been placed in unfamiliar cage environments. This mouse can mimic a FNE phenotype of humans and zolpidem and raclopride may be useful drugs to prevent acute insomnia including FNE.

8. Suzuki A, Sinton CM, Greene RW, Yanagisawa M. (2013) *Proc. Natl. Acad. Sci.* USA **110**(25): 10288-10293. Behavioral and biochemical dissociation of arousal and homeostatic sleep need influenced by prior wakeful experience in mice.

   There is a simple positive correlation between sleep need and sleepiness, although a detailed mechanism behind this relationship is still unclear. The authors found that sleepiness and sleep need can be regulated independently, or rather, dissociated from one another. In addition, through a phosphoproteomic approach, separate CNS (central nervous system) biochemical signatures associated with these behavioral parameters were identified.

9. Wolburg H, Mogk S, Scker S, Frey C, Meinert M, Schonfeld C, Lazarus M, Urade Y, Kubota BK, Duszenko M. (2012) *PLoS One* **7**(3):e34304. Late stage infection in sleeping sickness.

   Trypanosomes are the causative agent of sleeping sickness. However, no definitive proof of how the parasites reach and affect the brain has been presented. The authors showed that trypanosomes do not colonize the brain but reside near or within the glia limitans, from where they can re-populate blood vessels and disrupt the sleep wake cycles.

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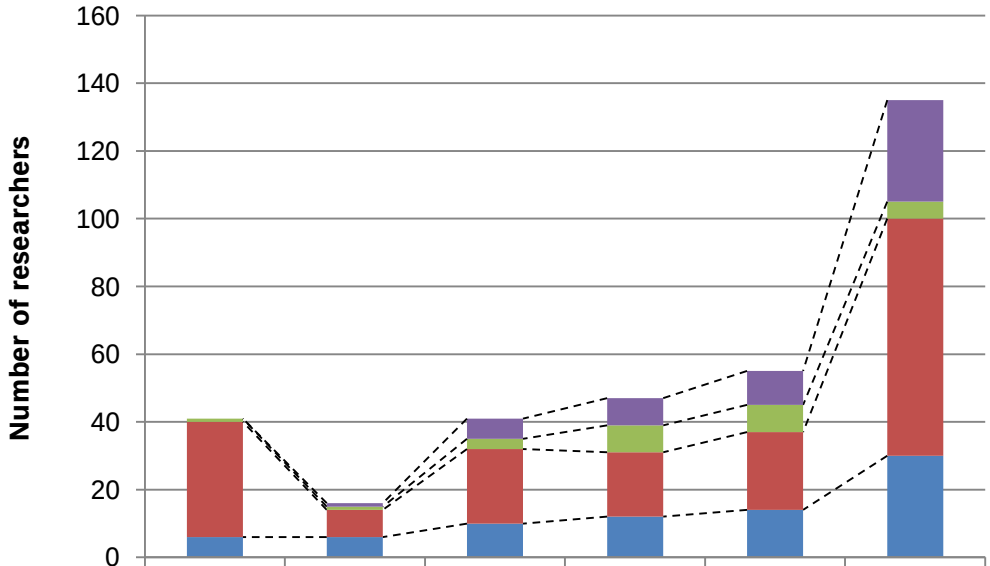
Appendix 4-2. Number of overseas researchers and annual transition

*Make a graph of the transition in the number of overseas researchers since the application.

Annual Transition in the Number of Overseas Researchers



	At beginning	FY2012	FY2013	FY2014	FY2015	Final goal
Researchers from abroad excluding PIs	0	1	6	8	10	30
PIs from abroad	1	1	3	8	8	5
Japanese researchers excluding PIs	34	8	22	19	23	70
Japanese PIs	6	6	10	12	14	30

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Appendix 4-3. Postdoctoral positions through open international solicitations

- In the “number of applications” and “number of selection” columns, put the number and percentage of overseas researchers in the < > brackets.

FY	Number of Applications	Number of Selection
FY2012	3 <1, 33%>	3 <1, 33%>
FY2013	169 <158, 93%>	12 <4, 33%>
FY2014	123 <144, 93%>	9 <3, 33%>
FY2015	98 <97, 99%>	4 <3, 75%>

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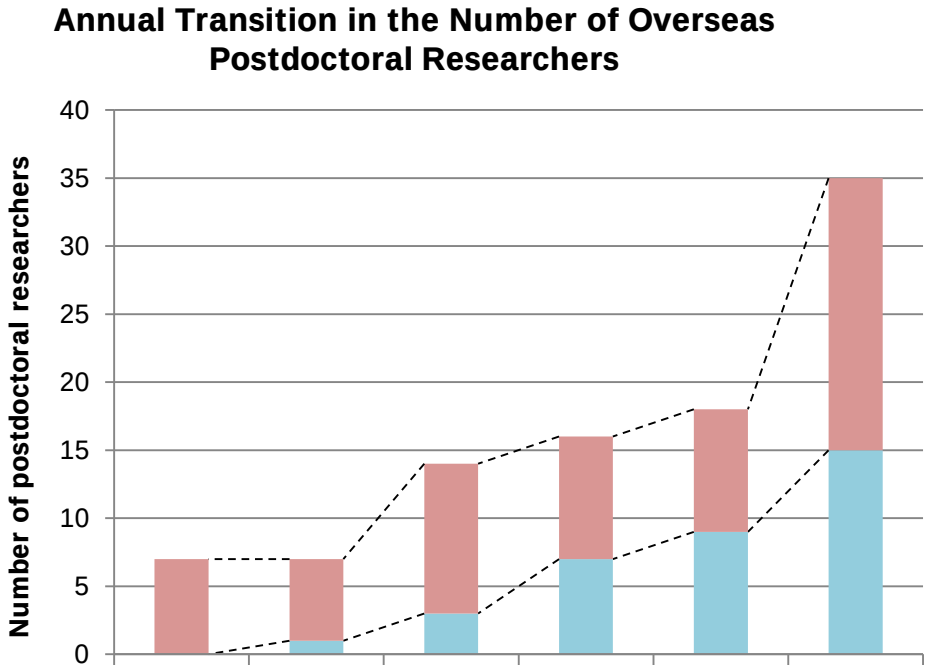
Appendix 4-4. Number of overseas postdoctoral researchers

* Make a graph of the transition in the number of overseas postdoctoral researchers since the project application was submitted.



	At beginning	FY2012	FY2013	FY2014	FY2015	Final goal
Japanese postdoctoral researchers	7	6	11	9	9	20
Overseas postdoctoral researchers	0	1	3	7	9	15

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Appendix 4-5. Status of postdoctoral researchers' employment at institutions

- List each researcher in 1 line. If the list exceeds this form, please add extra pages.

Period of project participation	Previous Affiliation (organization, *country)	Next Affiliation (Position title, organization, *country)	Nationality
2013.2.1-2013.3.31	RIKEN・Japan	Associate Professor・University of Tsukuba・Japan	Japan
2013.2.1-2013.8.31	University of Tsukuba・Japan	Research Administrator・University of Tsukuba ・Japan	Japan
2013.3.1-2013.12.31	Osaka Bioscience Institute・Japan	Researcher・Indiana University・USA	India
2013.4.1-2013.11.30	GreenSogna, Inc.・Japan	Unknown	Japan
2013.4.1-Present	Harvard University・USA		Japan
2013.4.1-Present	Osaka Bioscience Institute・Japan		Japan
2013.4.1-2013.11.30	Kanazawa University・Japan	Assistant professor・University of Tsukuba・Japan	Japan
2013.7.1-2014.6.30	University of Texas・USA	Appointed Assistant Professor・University of Tokyo・Japan	Japan
2013.7.1-Present	Osaka Bioscience Institute・Japan		France
2013.7.1-2014.9.30	RIKEN・Japan	Assistant Professor・University of Tsukuba ・Japan	Canada
2013.8.16-2013.11.30	Okayama University・Japan	Assistant Professor・University of Tsukuba ・Japan	Japan
2013.9.1-2013.11.30	Takeda Pharmaceutical Co.,Ltd.・Japan	Associate Professor・University of Tsukuba・Japan	Japan
2013.10.1-Present	Osaka Bioscience Institute・Japan		Japan
2014.2.16-Present	University of Basel・Switzerland		Germany
2014.3.1-Present	Friedrich Miescher Institute・Switzerland		Canada
2014.4.1-Present	Osaka University・Japan		Russia
2014.4.1-Present	University of Tsukuba・Japan		Japan
2014.4.1-Present	University of Tsukuba・Japan		Japan
2014.4.1-Present	University of Tsukuba・Japan		Japan
2014.4.1-2015.3.31	University of Tsukuba・Japan	Research Fellow of the JSPS(PD)・Japan	Japan
2014.4.1-Present	University of Tsukuba・Japan		Japan

Period of project participation	Previous Affiliation (organization, *country)	Next Affiliation (Position title, organization, *country)	Nationality
2014.6.16-Present	University of Texas・USA		Japan
2014.9.1-Present	Indiana University・USA		India
2014.9.16-2015.9.30	Max Planck Institute of Psychiatry・Germany	JSPS Postdoctoral Fellowship for Overseas Researcher・Japan	India
2015.4.1-Present	University of Tsukuba・Japan		England
2015.4.1-Present	University of Tsukuba・Japan		Japan
2015.5.1-Present	University of Texas・USA		China
2015.8.1-Present	University of Texas・USA		China

* The country in which the organization is physically located.

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Appendix 4-6. Holding International Research Meetings

For each fiscal year, indicate the number of international research conferences or symposiums held and give up to two examples of the most representative ones using the table below.

FY 2012-2013: 2 meetings

Major examples (meeting title and place held)	Number of participants
Title: The 1st Annual IIIS Symposium Venue: International Congress Center, Tsukuba	From domestic institutions: 169 From overseas institutions: 30
Title: The 2nd Annual IIIS Symposium Venue: International Congress Center, Tsukuba	From domestic institutions: 121 From overseas institutions: 22

FY 2014: 3 meetings

Major examples (meeting title and place held)	Number of participants
Title: “Homeodynamics in Clock, Sleep and Metabolism” (The 3rd Annual IIIS Symposium) Venue: University of Tokyo	From domestic institutions: 215 From overseas institutions: 17
Title: The 68th Fujihara Seminar Venue: IBM Amagi Homestead	From domestic institutions: 39 From overseas institutions: 24

FY 2015: 1 meeting

Major examples (meeting title and place held)	Number of participants
Title: The 4th Annual IIIS Symposium Venue: IIIS Building, University of Tsukuba	From domestic institutions: 126 From overseas institutions: 56

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Appendix 5-1. Host institution's commitment

1. In-kind contributions from host institution

(personnel, laboratory space, etc.)

Other resources provided by University of Tsukuba as support for IIIS

University of Tsukuba have provided IIIS with various resources as support. The provided support is equal to or greater than the support planned in the application for the WPI program as following;

1) University of Tsukuba established the Organization for the Support and Development of Strategic Initiatives, and IIIS received 10 million JPY in management expenses grants as support from the initiative.
2) Support was launched for research funding and also with applications for competitive funding.
3) Support the part of the personnel cost of Vice Center Director, Sakurai.
4) Measures were taken to provide personnel costs for 3 administrative positions for university personnel engaging in the key areas of general affairs and accounting. From July, 2015, arranged one URA to the research strategy and management team.
5) In FY2015 University support 70 million yen as the expenses for utility of the new research building from the indirect cost though university collected 50 million yen as the rental fee for own funding area of 2,000 m^2.
6) Support was provided for the research spaces of the Health and Medical Science Innovation Laboratory, University of Tsukuba Hospital E Building, Project Research Building and TARA Center, providing 5,000 square meters divided among the listed venues until the completion of relocation to the new research building in August 2015.

Tenure of PI

University of Tsukuba and IIIS will start the discussion on the tenure of PIs who produces sufficient research achievements using the reallocation system so that IIIS can remain "a globally unrivaled research center" beyond the end of the WPI program implementation period.

2. System under which the center's director is able to make substantive personnel and budget allocation decisions

Division of authority

All key matters concerning the operation in the institute have been decided by a top-down system of the Director as shown in 5-1.

Principal Investigators' meeting (PI meeting)

According to the establishment of IIIS, led by the administrative department, a PI meeting was established, during which the PI regularly submits opinions to the Director to determine important matters concerning the IIIS as shown in 5-1.

IIIS personnel committee

IIIS established a personnel committee to improve the way of researcher's appointment as shown in 5-1.

Introduction of a system to evaluate research results and ability-linked salary system

From FY2015 at University of Tsukuba, a quantitative evaluation index concerning research performance in terms of published papers and writings, granted external funding, and research alliance with public and for-profit organizations, is being established. We are thus considering the index as an evaluation tool to build a system of merit-based compensation as shown in 5-3.

3. Support for the center director in coordinating with other departments at host institution when recruiting researchers, while giving reasonable regard to the educational and research activities of those departments

Teaching qualification

Ten with Comprehensive Human Science Biomedical Sciences major (doctoral programs), 9 with Comprehensive Human Science Medical Sciences major (master's programs), 9 with Ph. D. Program in Human Biology (HBP) and one with Pure and Applied Sciences Chemistry major have a teaching qualification as shown in 5-5-1.

4. Revamping host institution's internal systems to allow introducing of new management methods

(e.g., English-language environment, merit-based pay, cross appointment, top-down decision making unfettered by conventional modes of operation)

Constitution of the administrative department

Administrative department is operated by the following four teams (General affairs, Accounting, Research strategy and management, and Alliance & Communication) led by the Administrative Director as shown in 5-2.

Use of English as official language and employment of bilingual staff

English is used as an official language at IIIS. A bilingual environment has been implemented, with 60% of administrative staff members fluent in spoken and written English as shown in 5-2.

Joint appointment system

With the purpose of enabling the Director to occupy concurrent posts at University of Tsukuba and UTSW, the joint appointment system was newly introduced to University of Tsukuba in March 2014 as shown in 5-3.

5. Utilities and other infrastructure support provided by host institution.

(facilities, e.g., laboratory space; equipment; land, etc.)

Cost of constructing a new research building

The Sleep Medicine Research Building costed approximately 3.8 billion yen and the expenses beyond those covered by the facility development subsidies of 2 billion yen kindly provided by MEXT were supported by in-house funding as shown in 5-4.

6. Support for other types of assistance

Tsukuba Short-term Study Program (TSSP)

As for the short-term training, as shown in the following table, we have already accepted many trainees using TSSP.

	Social position/duty position	Instructor	Nationality	Date of start of acceptance	Visit duration (days)
1	Medical school student	Masanori Sakaguchi	Taiwanese	July 7, 2014	54
2	Graduate (doctoral)	Lazarus Michael	Chinese	August29, 2014	6
3	Technical staff	Qinghua Liu	Japanese	June 1, 2015	61
4	Medical school student	Kaspar Vogt/Masashi Yanagisawa	American	May 25, 2015	68
5	Medical school student	Yu Hayashi	British	July 20, 2015	365
6	Graduate (master's)	Masanori Sakaguchi	Indian	June 2, 2015	89
7	Medical school student	Masanori Sakaguchi	Taiwanese	July 14, 2015	55
8	Medical school student	Masanori Sakaguchi	Taiwanese	July 14, 2015	55
9	Graduate (doctoral)	Masanori Sakaguchi	Thai	January 18, 2016	90
10	Graduate (master's)	Lazarus Michael	German	January 4, 2016	61

World Premier International Research Center Initiative (WPI)

Appendix 5-2. Female researchers

*Enter the number and percentage of female researchers in the top of each space and the total number of all the researchers in the bottom.

		FY2012	FY2013	FY2014	FY2015	Final goal
Researchers		6, 38%	12, 29%	13, 28%	15, 28%	35, 30%
		16	41	46	54	115
	Principal investigators	0, 0%	0, 0%	2, 10%	2, 9%	1, 7%
		7	13	20	22	15
	Other researchers	6, 67%	12, 43%	11, 42%	13, 41%	34, 34%
		9	28	26	32	100