National University Corporation
Tokyo Medical and Dental University

# Faculty of Medicine

# Imperial College London
# and
# TMDU
# Exchange Programme 2012
# for
# In-coming students

**June 2011**

Educational Affairs Section
Student Affairs Department
Tokyo Medical and Dental University
1-5-45 Yushima, Bunkyo-ku, Tokyo
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Japan

## 1. Introduction

This Course guidebook is designed for the exchange students who are going to undertake their BSc projects at Tokyo Medical and Dental University, which is cooperated as the Exchange Student Programme by Imperial College London and Tokyo Medical and Dental University ("TMDU"). Therefore, the projects listed in this course guide are available only for the selected students of this exchange programme.

## 2. Schedule

We expect you arrive in Tokyo on 25th or 26th of February 2012. TMDU Project will commence on 29th of February, 2012. You would be expected to start your project and work your lab on the same day as the project starts. You are working on the project from 29nd February to 3th April (5weeks) and have Easter Break from 4th April to 10th April (1 week). After Easter Break, you are working on your project from 11th April to 11th May for 4 weeks and 3 days and write-up for a week.

For preparing your project, I strongly recommend you contact your expected supervisor and talk them about the details of your project before your arrival. Some time, the circumstance of the laboratory might change when you do not have any reply from him/her, please ask Mr Alekseiv or send e-mail to the following address : m.sugawara.adm@cmn.tmd.ac.jp There might be a possibility to change in the content of your project due to the time limitation for the project.

**In-coming TMDU - Course Map 2012**

| | | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 25-26/02/2012 | Mon 27 Feb - Tue 28 Feb 2012 | 29-Feb-12 | 5-Mar-12 | 12-Mar-12 | 19-Mar-12 | 26-Mar-12 | 02-03/04/2012 | 4-10/04/2012 | 11-13/04/2012 | 16-Apr-12 | 23-Apr-12 | 30-Apr-12 | 7-May-12 | 14-May-12 | 19-20-May-12 | 21-May-12 | 28-May-12 | 4-Jun-12 | 11-Jun-12 |
| Week beginning 27 February (Monday) 2012 | 21 | 22 | 22 | 23 | 24 | 25 | 26 | 27 | | 28 | 29 | 30 | 31 | 32 | 33 | | 34 | 35 | 36 | 37 |
| | | | Project I (pre-Easter) - 5 wks | | | | | | | Project II (post-Easter) - 4 wks & 3 days | | | | | | | | | | |
| Year 4 (BSc) In-coming TMDU Exchange Students | Arrival | Induction | Project - 5 weeks | | | | | | Easter Break (1 week) | Project cont'd - 4 weeks and 3 days | | | | | Write-up | Return | Oral Presentation / Submission of write-up | Mark submission by Fri 1 June | | BSc Vivas |
| | 25-26/02/2012 | Mon 27 Feb - Tue 28 Feb 2012 | 29-Feb-12 | 5-Mar-12 | 12-Mar-12 | 19-Mar-12 | 26-Mar-12 | 02-03/04/2012 | 4-10/04/2012 | 11-Apr-12 | 16-Apr-12 | 23-Apr-12 | 30-Apr-12 | 7-May-12 | 14-May-12 | 19-20-May-12 | 21-May-12 | 28-May-12 | 4-Jun-12 | 11-Jun-12 |
| | 21 | 22 | 22 | 23 | 24 | 25 | 26 | 27 | | 28 | 29 | 30 | 31 | 32 | 33 | | 34 | 35 | 36 | 37 |

**In-coming Exchange Students: Dates**

- **End of Part B exams ; 23 Feb (Thursday) 2012**
- **Arrival at TMDU, Tokyo, Japan: Sat 25 Feb - Sun 26 Feb 2012**
- **Induction at TMDU: Mon 27 Feb – Tue 28 Feb 2012**
- **Project (Part I – Before the Easter Break) – 5 weeks:**
  **Wed 29 Feb – Tue 3 April 2012**
- **Easter Break: Wed 4 April – Tue 10 April 2012**
- **Project (Part II – After the Easter Break) - 4 weeks & 3 days:**
  **Wed 11 April – Fri 11 May 2012**
- **Write-up*: Monday 14 May – Friday 18 May 2012**
- **Return to Imperial College, London, UK: 19 - 20 May (Saturday/Sunday) 2012**
- **Oral Presentation of Project : Monday 21 – Thursday 24 May 2012**
- **Write-up submission : Thursday 24 May 2012 -2pm**

***Note: In some BScs, the oral presentation of project may be on Thursday 17 or Friday 18 May 2012 – Students should check with the BSc Course Director(s) before booking a return flight and arrange their presentation to be held between Monday 21 – Thursday 24 May 2012, if possible.**

## 3. Application form (Sample)

This form is only for the use of the Exchange Programme between Imperial College and TMDU. This form and all the information regarding this exchange programme will be available from the Student Placement Office (Mr Antony Aleksiev, Curriculum Administrator (BSc), Undergraduate Medicine Office:( a.aleksiev@imperial.ac.uk) at Imperial College London. The completed application form will be returned to Mr Aleksiev. Mr Aleksiev will take all the procedure for this exchange programme at Imperial College London.

**Faculty of Medicine Tokyo Medical and Dental University**

# Application Form for Exchange Student 2009

## 1. Personal Information

Family Name: _____ All Other Name: _____

Title: _____ Date of Birth: _____ (Month/ Day/ Year)

Sex: ☐male ☐female Marital Status: ☐married ☐unmarried

Country of Birth: _____ Nationality: _____

Home address: _____

_____

Phone Number: _____ Email Address: _____

Name and Address of Parent(s), guardian(s) or next of kin:

_____

_____

Special Dietary Needs: _____

Please state any medication you are taking: _____

_____

## 2. Project Details

Period of stay in Japan for the Project: from _____ to _____

Title of Project in TMDU : _____

_____

Name of TMDU Supervisor: _____

Name of Home Supervisor: _____

## 3. Academic Qualifications

High School

| from | to (Month / Year) | |
|---|---|---|
| ___/___ | ___/___ | _____ |

Colleges, Universities

| | | |
|---|---|---|
| ___/___ | ___/___ | _____ |
| ___/___ | ___/___ | _____ |

## 4. Accommodation

You will receive an application form for your accommodation, named “International Student House” located in Kohnodai, Ichikawa-city, Chiba-prefecture after your application procedure completed. Chiba locates next Tokyo. You will work on your project in Yushima-campus (Main Campus), which locates in Tokyo. It takes an hour from your accommodation to Yushima-campus. (http://www.tmd.ac.jp/english/outline/access/index.html).

It takes about 40min on foot from your accommodation to the nearest station (JR Ichikawa-station). If you take a bus (Keisei Bus), it will take 15-20min. At JR Ichikawa-station, you change from bus to train and then catch a train (JR Soubu Line) to JR Ochanomizu Station.

- **Accommodation Fee**

You will be required to pay for the accommodation fee after your arrival.

・How much would it be?

**Monthly payment：** Room charge (single room): Yen 5,900 per month

We do not calculate the room charge by the day you actually stay in the accommodation. Therefore, you will be required to pay for 4 months (February, March, April and May).

・Where you pay?

1st floor of the Building 1 West

(http://www.tmd.ac.jp/english/outline/index.html)

Also you must pay for Maintenance charge: Yen 500 per month. You pay for these fees at the administration office (1st floor) in International Student Hall.

- **The other charges**

・Clean-up after your move-out: Yen 19,000

(Administration Office in International Student Hall)

・Bed rental for three months: Yen 8,000

(Including mattress, sheets, pillow, pillowcase, duvet, duvet cover, blanket)

- Internet (optional)

The internet service is optional. Therefore, if you would like to take this service, please send me e-mail (m.sugawara.adm@cmn.tmd.ac.jp). The monthly charge would be Yen2730. You are required to pay in advance and must prepay for the next month before the 25th of every month. If you forget to pay the charge for the following month by the due date, your contact is automatically cancelled. There is difficulty for you to face. You cannot transfer money from your bank into the bank account the internet company ask you to pay in as long as your bank is not Japanese one. Therefore, you may pay for the usage of the first month after your arrival.

The approximate total amount of costs for your accommodation is around Yen 64,720 plus TAX for three months. (this cost does not including the utility fees)

**N.B. Utility fees such as Gas, Electricity, Water and Internet (optional) are not included in the room charge. You will be sent the bills for Gas and Electricity by the companies and can pay at Convenience store and post office. However, you will be sent the bills for water by Administration office in your residence and you pay for the fee there.**

**Please do read the rule provided by the residence and keep the rule during your stay.**

### 6. Health Insurance

When you have a minor illness, you can walk in Health Service Centre of TMDU without appointment. You will be given a prescription. For the treatment in the centre, you will not be charged. However, you are in case of a more serious illness, you can go to our affiliated hospital or any other hospitals. However, you will be charged for any treatments by the staff at our affiliated hospital and at any hospital. Therefore, I strongly recommend you to purchase health insurance before your travel.

## 7. Messege from the 2011 exchange students

**Miss Kelly Ameneshoa**
**Department :** Department of Molecular Virology (Virology)
**Supervior;** Professor Shoji Yamaoka

The TMDU exchange programme is a fantastic opportunity for medical students and the three months I have spent in Japan have been the quickest of my life. I was investigating HIV host restriction factors in the molecular virology laboratory. I had no previous laboratory experience but this was no obstacle. Different lab members took the time to train me in the many techniques I used and both my supervisor and professor were really supportive throughout the project. Beyond work, my colleagues were unbelievable welcoming and many have become good friends. Be prepared for lots of invitations for welcome parties, drinks, dinner etc! During the exchange we also had the opportunity to meet and get to know many of the medical students at TMDU (including students who had been/are going to Imperial College for their exchange).

Outside TMDU, living in Japan was an incredible experience. It is a beautiful country with something to offer everyone. I went travelling for two weeks and experienced more rural Japan as well as the city life outside Tokyo. After the obvious places such as Kyoto, I would definitely recommend a trip to Kanbayashi to see the snow monkeys and stay in the ryokan nearby. The accommodation at Ichikawa is small but functional. Many international TMDU students stay there so it is also a great chance to meet more people. The accommodation also offer free bicycle rental which is very convenient for travelling to the train station and avoids the bus fare.
I cannot recommend the TMDU exchange programme enough; it is one of the best experiences I have had and one that will never leave me

**Mr Andrew Gordon**
**Department :** Department of Surgical Oncology
**Supervior;** Professor Kenichi Sugihara and Associate Professor Hiroyuki Uetake

In the three months we spent in Japan, there was never a dull moment. From our arrival one sunny February morning, every day was filled with excitement and interest. Not once did I question my decision to apply for the programme.

I spent my time researching into Gastric cancer in the department of Surgical Oncology. In keeping with all my encounters in Japan, the staff were extremely helpful and friendly. It was fascinating to observe and work with surgeons at the cutting edge of their field. The chance to learn from such innovative people was priceless. Other than the research, the university has many extra-curricular activities which are very accessible and great fun.

But the attraction of this exchange extends past it's unquestionable academic benefits. In addition, you're able to sample the truly unique way of life in Japan. The value of this cannot be emphasised enough. Aside from the incredible atmosphere of your day-to-day life in the vibrant capital, the rest of the country provides a diverse and exquisite range of opportunities. From the endless beauty and tranquility in Kyoto or Nara, to the stunning views in the Mount Fuji area, irrespective of one's preferences, you will be blown away by what's on offer. Perhaps most impressive is the sublime combination of a traditional Japanese hotel (Ryokan) and hot spring (Onsen). After overcoming the 'difference' in bathing etiquette, the relaxing nature of the baths will soon set in and give you a chance to unwind, before the sumptuous 'Kaiseki' dinner which follows.

The setup at TMDU works exceedingly well, with a well-developed infrastructure. Despite the tragic earthquake and anxiety of living through a major natural disaster, my time in Japan was unforgettable and will undoubtedly have a positive impact on my future. I would definitely recommend this phenomenal experience to anyone on the medical course.

2011 Imperial College exchange students with ex-Dean Ohno Faculty of Medicine (Center), Prof. Tanaka (first from right), Assoc. Prof. Takada (second from left) and their supervisiors at the beging of their stay.

2011 Imperial College exchange students with President Ohoyama (center of the second line), Assoc. Prof. Takada(right of the sencond line) and Ms. Sugawara (TMDU programme coordinator, left of the second line)at the end of their stay.

※ Regarding the subjects offered by each department, there is a possibility to be changed in the contents of the project due to the short time of your project period.

## *01 Department of Cell Biology*

**Project title:** Development of a new light-activated signaling molecule and manipulation of cellular functions

**Supervisor:** Prof. Takao Nakata – nakata.cbio@tmd.ac.jp

**Key words**: cell signaling, synthetic biology, caged protein, molecular biology, protein structure and function

**Background**: Thanks to the GFP technology, in last decades, we could observe dynamics of proteins, as well as regional activity of enzymes within cells. Some of them were really amazing, but we are now not satisfied to use light for simple observation, and are willing to manipulate intracellular signaling by using light. There are several natural light-activated signaling molecules, but they are rare case. We make chimera proteins of light sensitive proteins and signal proteins to make novel signaling protein, and we use it to analyze the cellular function of the signal protein quantitatively with high spatiotemporal resolution.

**Subject**: Exchange students will challenge to make a novel light-induced protein, using molecular biology techniques and structural biology information. Once she or he successfully makes the chimera protein, she or he introduces it to the cultured cells (neurons) and performs cell biological analysis.

## *02 Department of Neuroanatomy and Cellular Neurobiology*

**Project title:** Cell biological analysis of morphological dynamics in neurons

**Supervisor:** Prof. Sumio Terada – terada.nana@tmd.ac.jp

**Key words:** cytoskeleton, small G protein, neuron, intracellular transport, microscopy

**Subject #1: Screening and identification of novel functional molecules influencing cytoskeletal dynamics in neurons**

We are now developing a new experimental system for screening functional molecules that influence cytoskeletal dynamics in neurons. Candidate proteins responsible for morphological changes will be studied extensively by various molecular and cell biological experiments.

**Subject #2: Studies on neuron-specific small G protein function in neuronal morphology**

We have found that some of the neuron-specific small G proteins play distinctive roles in neuronal morphology. We are currently studying their molecular mechanism that regulates neuronal shape, by using cell-lines, primary cultured neurons and molecular biological methods.

**Animal Experiment:** Concerning Subject #1, no animal experiments are planned, and only cell biological and molecular biological experiments using established cell-lines are considered. Concerning Subject #2, we usually use mice for primary cultured neurons.

## *03 Department of Systems Neurophysiology*

**Project title:** Organization of axonal projections in the cerebellum

**Supervisor:** Prof. Izumi Sugihara – isugihara.phy1@tmd.ac.jp

**Key words:** light microscopy; rodents; neuronal labelling; axonal reconstruction; cerebellum

**Background:** Axons allow nerve cells (neurons) to signal each other over long distances. Axonal projection patterns determine the organization of the central nervous system. Morphological techniques to analyse axonal projections include tracing the trajectory of labelled axons. We developed a computer-aided light microscopy system with the camera lucida apparatus for this purpose. We have been studying afferent and efferent axonal connections in the cerebellum with this system (Refs. 1-4), since they are essential in understanding the cerebellar function.

**Subject:**We propose you to study the axonal projection pattern from the vestibular nucleus to the cerebellum, which still remains unknown. This projection conveys vestibular information to the cerebellum and is essential in the cerebellar control of locomotion, posture and eye movements. A microinjection of the neuronal tracer to the vestibular nucleus in anesthetized rodents can label vestibuloocerebellar single axons. Labelled axons can be reconstructed completely in three-dimensional space. The branching pattern of reconstructed axons will be compared with the striped molecular expression pattern of the cerebellum (Ref. 5).

**References:**

1. Figure 20-20 (page 512) of Nolte J. The Human Brain, An Introduction to its Functional Anatomy, 6th ed. Mosby Elsevier, 2009.
2. Figure 20.5 (page 814) of Nieuwenhuys R, Voogd J, van Huijzen C. The Human Central Nervous System, 4th ed. Springer, 2008.
3. Figure 20.17B,C (page 487) of Squire LR et al. Fundamental Neuroscience, 3rd ed. Academic Press, 2008.
4. Figure 14 (page 222) in Chapter 3 of Paxinos G ed. The Rat Nervous System, 3rd ed. Elsevier Academic, 2004.
5. Figure 9-20C (page 184) of Koeppen BM, Stanton BA ed. Berne & Levy Physiology, 6th ed. Mosby Elsevier, 2010.

## *04 Department of Physiology and Cell Biology*

**Project title:** Molecular mechanism and physiological role of autophagy

**Supervisor:** Prof. Noboru Mizushima – nmizu.phy2@tmd.ac.jp

**Key words:** Autophagy, protein degradation, metabolism, nutrition, amino acids, starvation, insulin signaling

**Subject:**Studies in our laboratory focus on autophagy, a dynamic degradation system within cells. Eukaryotes have two major protein degradation systems: one is the ubiquitin-proteasome system that accounts for the selective degradation of most short-lived proteins, and the other one is the lysosomal system. Autophagy is the primary means for the degradation of cytoplasmic constituents in the lysosome. We have dissected the autophagic process in mammalian cells at the molecular level, and are currently analyzing several novel factors involved in autophagy and the nutrient signaling.

We also study the physiological role of autophagy using an autophagy-indicator mouse model (GFP-LC3 mice) and autophagy gene-knockout mice. We have shown that autophagy is upregulated following food withdrawal and during early embryogenesis and neonatal starvation period in mice. We proposed that “induced autophagy” is important for intracellular generation of amino acids by breakdown of self proteins during these processes. On the other hand, we showed that basal autophagy has a critical role in intracellular protein quality control under normal conditions, which is important for prevention of neurodegeneration and spontaneous tumorigenesis. We are now examining additional roles of autophagy using new mouse models in which autophagy is perturbed in many other tissues.

**Animal Experiment:** We use mice for *in vivo* experiments, which include scarification and starvation treatment.

**References**

Takamura, A., Komatsu, M., Hara, T., Sakamoto, A., Kishi, C., Waguri, S., Eishi, Y., Hino, O., Tanaka, K., Mizushima, N. Autophagy-deficient mice develop multiple liver tumors. ***Genes Dev.*** 25: 795-800 (2011).

Yoshii, S.R., Kishi, C., Ishihara, N., Mizushima, N. Parkin mediates proteasome-dependent protein degradation and rupture of the outer mitochondrial membrane. ***J. Biol. Chem.*** in press

Hosokawa, N. et al. Nutrient-dependent mTORC1 association with the ULK1-Atg13-FIP200 complex required for autophagy. ***Mol. Biol. Cell*** 20: 1981-1991 (2009)

Mizushima, N. et al. Autophagy fights disease through cellular self-digestion ***Nature*** 451:1069-1075 (2008)

Tsukamoto, S. et al. Autophagy is essential for preimplantation development of mouse embryos. ***Science*** 321: 117-120 (2008)

Hara, T. et al. FIP200, a ULK-interacting protein, is required for autophagosome formation in mammalian cells. ***J. Cell Biol.*** 181: 497-510 (2008)
Hara, T. et al. Suppression of basal autophagy in neural cells causes neurodegenerative disease in mice. ***Nature*** 441, 885-889 (2006).
Kuma, A. et al. The role of autophagy during the early neonatal starvation period. ***Nature.*** 432, 1032-1036 (2004).

## *05 Department of Medical Biochemistry*

**Project title:** Regulation and function of mammalian Hippo pathway and RASSF

**Supervisor:** Prof. Yutaka Hata yuhammch@tmd.ac.jp
http://www.tmd.ac.jp/english/mbc/index.html

**Subject:** Our main focus is on the Hippo pathway and RASSF proteins. The Hippo pathway is a newly emerging signaling pathway to regulate cell proliferation and cell death. It is a kinase cascade to phosphorylate and negatively regulate transcription co-activators named YAP and TAZ. The co-activators enhance the transcriptions of genes that promote cell cycle and inhibit cell death. The loss of function of the Hippo pathway leads to tumorigenesis. The dysfunction of the Hippo pathway is frequently detected in human cancers (for instance, 60% in breast cancer and 70% in astrocytoma). As YAP and TAZ induce epithelial-mesenchymal transition and the expression of stem cell markers in cancer cells, the impairment of the Hippo pathway causes metastasis and resistance against cancer therapies, and correlates with poor prognosis. RASSF is a family of proteins that have Ras-association (RA) domains. Human genome harbors ten RASSF genes, RASSF1 to RASSF10. RASSF1 to RASSF6 have the RA domain in the middle region and form a subfamily named the classical RASSF, while RASSF7 to RASSF10 have the RA domain in the N-terminus. Recent studies including ours have revealed that the Hippo pathway and the classical RASSF are closely related to each other. The classical RASSF proteins induce cell cycle arrest and apoptosis and suppress inflammatory signaling. The trigger to activate the Hippo pathway simultaneously induces the RASSF-dependent biological events. Thereby, the Hippo pathway and RASSF proteins co-operate to prevent tumorigenesis. The loss of RASSF proteins is common in human cancers. RASSF1A (a splicing variant of RASSF1) and RASSF6 are suppressed in 50% of lung cancer and in 90% of childhood B-cell acute lympocytic leukemia, respectively. The importance of the Hippo pathway and RASSF proteins is not only limited in the field of cancer biology. YAP and TAZ regulate adipogenesis, osteogenesis, and myogenesis. RASSF proteins are implicated in inflammatory diseases. All these findings support the significance of the Hippo pathway and RASSF proteins in human diseases.

We are studying to examine how the disorders of Hippo pathway and RASSF proteins cause tumorigenesis at the cell level and at the animal level. We search for the chemical compounds to inhibit and stimulate the Hippo pathway and to antagonize the function of YAP and TAZ. These compounds facilitate the study of the Hippo pathway and RASSF proteins. They may be therapeutically useful.

*We use mice, rats, and rabbits in the laboratory. However, it is possible to plan a study without using animals.

## *06 Department of Immune Regulation*

**Project title:** “Molecular Pathogenesis of Allergy and Immunodeficiency”

**Supervisor:** Prof. Hajime Karasuyama – karasuyama.mbch@tmd.ac.jp

**Key words:** basophils, allergy, inflammation, parasitic infections, primary immunodeficiency, Hyper-IgE syndrome, animal models

**Subject #1: Emerging roles for basophils in protective immunity and allergy**

Basophils are the least common granulocytes, and represent less than 1% of peripheral blood leukocytes. Although basophils were first documented by Paul Ehrlich more than 120 years ago, their functional significance has remained an enigma for a long time. We have recently succeeded in generating novel tools suitable for analysis of basophil functions, namely a basophil-depleting antibody, engineered mice for selective and inducible ablation of basophils in vivo, and engineered mice expressing green fluorescence protein only in basophils. Taking advantage of these tools, we identified previously unrecognized, non-redundant roles for basophils *in vivo*. Basophils are crucially involved in the acquisition of resistance against repeated infections with parasites, including blood-sucking ticks and intestinal helminths. Basophils also play pivotal roles in the development of allergic disorders, such as IgG-mediated systemic anaphylaxis and IgE-mediated cutaneous allergic inflammation. The physiological and pathological roles of basophils will be further clarified.

**References**

1. Karasuyama, H. et al.: Nonredundant roles of basophils in immunity. ***Annu. Rev. Immunol***. 29: 45-69, 2011.
2. Karasuyama, H., et al.: Emerging roles of basophils in protective immunity against parasites. ***Trends Immunol***. 32: 125-130, 2011.
3. Karasuyama, H. et al.: Newly-discovered roles for basophils: a neglected minority gains new respect. ***Nat. Rev. Immunol***. 9: 9-13, 2009.

**Subject #2: Hyper-IgE syndrome: its causative genes and defects in multiple cytokine signals**

Hyper-IgE syndrome (HIES) is a compound primary immunodeficiency, characterized typically by recurrent staphylococcal infections, and severe atopic dermatitis with highly elevated serum IgE. Its etiology remained mysterious for more than 40 years, despite of extensive genetic approaches. We recently identified a homozygous mutation of the TYK2 gene in the autosomal-recessive form of HIES, and subsequently dominant negative mutations of the STAT3 gene in the sporadic and autosomal-dominant forms of HIES. Both gene products are known to be involved in the signal transduction of various cytokines. Indeed, the patient’s cells displayed defects in multiple cytokine signals, including IL-6, IL-10, and IL-23, which accounts for the patients’ complex clinical manifestations. We have recently

succeeded in establishing a mouse model of human HIES with the dominant-negative mutation of STAT3 to study more details of the molecular pathogenesis of HIES.

**References**


1. Saito, M. et al.: Defective IL-10 signaling in hyper-IgE syndrome results in impaired generation of tolerogenic dendritic cells and induced regulatory T cells. ***J. Exp. Med***. 208: 235-249, 2011.
2. Minegishi, Y. et al.: Molecular explanation for the contradiction between systemic Th17 defect and localized bacterial infection in hyper-IgE syndrome. ***J. Exp. Med***. 206: 1291-1301, 2009.
3. Minegishi, Y. et al.: Dominant-negative mutations in the DNA-binding domain of STAT3 cause hyper-IgE syndrome. ***Nature*** 448: 1058-1062, 2007.


(Laboratory mice including genetically engineered mice are examined in some experiments of both projects. However, experiments with no use of animals can be planned.)

## *07 Department of Immunotherapeutics*

**Project title:** Immunological understanding of virus-mediated diseases and approaches for immunotherapy

**Supervisor:** Prof. Mari Kannagi - kann.impt@tmd.ac.jp

**Key words:** Retrovirus, Pathogenesis, Host defense, T-cell immunity, Innate immunity, Immune suppression, Adult T-cell leukemia, AIDS.

**Subject:** We investigate the disease mechanisms of human retroviral infection, such as AIDS caused by human immunodeficiency virus type 1 (HIV-1) and adult T-cell leukemia caused by human T-cell leukemia virus type I (HTLV-I). Theses diseases are not simply explained by the direct pathogenic effects of the viruses, but influenced by a complex interplay between viruses and the host immune system. The aim of our research is the understanding disease mechanisms and the development of prophylactic and therapeutic strategies in these viruses infection.

**Specific research subjects:**

1. Immune tolerance in adult T-cell leukemia.
2. Molecular mechanism of HTLV-1 gene suppression.
3. Host factors required for HIV-1 replication especially for HIV-1 integrase.
4. Immunological suppression of HIV-1 replication.

(Animal experiments might be included in subjects #1 and #2, but dispensable.)

## *08 Department of Molecular Virology (Virology)*

**Project title:** Mechanism and therapeutic intervention of human retrovirus infection

**Supervisor:** Professor Shoji Yamaoka - shojmmb@tmd.ac.jp

Key words: human retroviruses, HIV-1, HTLV-I, virus replication, host factors, NF-kappaB, signal transduction.

**Subject #1: Studies on human retroviruses and host cell factors**
We mainly deal with retrovirus-mediated oncogenesis and immunodeficiency in humans; Human T-cell Leukemia Virus type 1 (HTLV-1) causing Adult T-cell Leukemia (ATL) and Human Immunodeficiency Virus type 1 (HIV-1) causing Acquired Immunodeficiency Syndrome (AIDS). HIV-1 has long been studied virologically, but little is known about cellular factors required for HIV-1 replication. Because HIV-1 rapidly undergoes mutations under current drug therapies, it is necessary to identify and target cellular factors that the virus requires for its replication. Genetic approaches coupled with an expression cloning are being undertaken to identify cellular proteins essential for HIV-1 replication.

**Subject #2: Mechanism of constitutive NF-kappaB activation in cancer cells**
One of the prominent features with ATL is constitutive activation of cellular transcription factors in leukemic cells even in the absence of viral antigens, and we are focusing on the NF-kappaB family of transcription factors which are pivotal for the survival and growth of HTLV-1-infected ATL cells. Accumulating evidence indicates that constitutive activation of NF-kappaB contributes to the manifestation of malignant phenotypes such as resistance to anti-cancer drugs, invasion and metastasis, but its molecular mechanism remains poorly understood. We previously demonstrated that hematopoietic tumours including Hodgkin lymphoma and ATL cells overexpress a serine-threonine kinase, NF-kappaB inducing kinase (NIK), which is responsible for constitutive NF-kappaB activity and related gene expression in these cancer cells. Studies are underway to elucidate the molecular mechanism of its overexpression. Lung, colon and breast cancer cells as well as Epstein-Barr virus infected human B-cells are also being studied for the mechanism(s) of constitutive NF-kappaB activation.

**Animal Experiments:** Students are expected to learn how to culture cells *in vitro*, infect them with retrovirus or lentivirus and analyse them with molecular biological techniques. Animal experiments are not planned in these subjects.

## *09 Department of Comprehensive Pathology*

**Project title:** Pathogenesis of haematological malignancies: regulatory mechanisms of proliferative/apoptotic signals

**Supervisor:** Prof. Masanobu Kitagawa **-** masa.pth2@tmd.ac.jp

**Key words:** haematological malignancies, bone marrow, lymph node, proliferation, apoptosis, oncogenesis, angiogenesis, real-time PCR, cell line

**Subject:** We study the molecular mechanisms of the regulation of proliferative/apoptotic signals in tumour cells from haematological malignancies. We can prepare the fresh samples and cDNA samples from the bone marrow/lymph node of normal individuals and cases with various haematological diseases such as acute leukaemia, myelodysplastic syndromes, multiple myeloma, and malignant lymphoma. We also have the samples of pre-treated and post-treated subjects of these cases. You can design the real-time PCR primers and probes of genes of your own interest to analyze the proliferative/apoptotic signals in tumour cells from haematological malignancies. You can also design the experiments using cell lines to confirm the effects of treatment in case samples on proliferative/apoptotic signals.

## *10 Department of Nephrology*

**Project title:** Ion transporter in kidney epithelial cell

**Supervisor:** Prof. Sei Sasaki **-** ssasaki.kid@tmd.ac.jp
Associate Prof. Shinichi Uchida - suchida.kid@tmd.ac.jp

**Key words:** ion channel, ion transporter, kidney epithelial cell, electrolytic disorder, blood pressure disorder, pseudohypoaldosteronism type II (PHA II), WNK4 (with no lysine (K)) kinase, thiazide-sensitive Na-Cl cotransporter (NCC), phosphorylation

**Subject #1: Disease causing mutant ion transporter in kidney epithelial cell**

We have worked on the mechanisms for genetic disease in the kidney. It is known that mutations in ion channel and transporter result in genetic diseases that cause electrolytic and blood pressure disorder, such as Bartter syndrome, Gitelman syndrome and Liddle syndrome. One of the mechanisms for these diseases is the mis-localization of the mutant transporter or channel in kidney tubules.

In this subject, we will make over-expression vectors of ion transporter that carries disease causing mutation and transfect them to kidney epithelial cells. We will check the cellular localization of the mutant in the cell, to see whether the mutation affect the localization of the transporter.

**Subject #2: Phosphorylation and localization of thiazide-sensitive Na-Cl cotransporter in kidney epithelial cell**

Recently, we are focusing on the mechanism of pseudohypoaldosteronism type II (PHA II), an autosomal-dominant disorder characterized by hyperkalemia and hypertension. We have reported that a missense mutation of WNK4 (with no lysine (K)) kinase increased function of thiazide-sensitive Na-Cl cotransporter (NCC) by changing phosphorylation status and localization in *vivo*. Phosphorylation of NCC on Ser71 was increased in the PHA II model mice.

In this subject, we will make over-expression vectors of phospho-mimicking and phospho-deficient NCC on several predicted phosphorylation site(s). Then, we will transfect them to kidney epithelial cell lines and check their localization in vitro, to see the effect of phosphorylation of each site on the cellular localization.

**Animal Experiments**: Students are expected to learn how to culture cells in vitro, transfect over-expression vector and analyze them with molecular biological techniques in both subjects. Animal experiments are not planned in our subjects.

**References**

1. Ohta A et al. Targeted disruption of the Wnk4 gene decreases phosphorylation of Na-Cl cotransporter, increases Na excretion and lowers blood pressure. Hum. Mol. Genet. 18:3978-86, 2009.

2. Noda Y et al. Reciprocal interaction with G-actin and tropomyosin is essential for aquaporin-2 trafficking. J. Cell Biol. 182:587-601, 2008.
3. Yang SS et al. Molecular pathogenesis of pseudohypoaldosteronism type II: generation and analysis of a Wnk4(D561A/+) knockin mouse model. Cell Metab. 5:331-344, 2007.
4. Sohara E et al. Pathogenesis and treatment of autosomal-dominant nephrogenic diabetes insipidus caused by an aquaporin 2 mutation. Proc Natl Acad Sci U S A. 103: 14217-14222, 2006.

### *11 Department of Life Science and Bioethics Research Center*

**Project title:**
Dive into vascular bilogy: Exciting Journey to the heart of cardiovascular disease.

**Supervisor:** Prof. Masayuki Yoshida - masavasc@tmd.ac.jp

**Key Words:** vascular biology, atherosclerosis, adhesion molecules, insulin resistance, chronic inflammation, innate immunity

**Subject #1. Cell adhesion in vascular diseases**
Inflammatory responses consists critical part of vascular diseases such as atherosclerosis. Numerous medications including ACEI, AT1 receptor blockers (ARB), statins have been shown to exhibit beneficial effects in these conditions. Our laboratory has been involved in interaction between leukocytes and vascular endothelium and its modulation. We have utilized a well established flow chamber model to examine adhesion assay in vitro. We recently developed a system to investigate cell adhesion in vivo using intravital microscopy (IVM). A student can learn and conduct some of these adhesion assay techniques under a hand-to-hand instruction by our staff.

**Subjects #2. Lipid abnormality and metabolic disorders**
Our laboratory focuse on lipid metabolism and its influence on inflammation in metabolic disorders including type 2 diabetes. Our current interests reside in a role of lipid absorption in metabolic disorders and its vascular consequences. We utilize a unique technique to directly monitor intesitnal absorption and its consequensial mesenteric lymph flow in situ. This technique enables us to examine potential roles of immuno-inflmmator cells at intesitinal mucosa and/or barterial flora during specific lipids and fatty acids nutrients. A student can learn and conduct these animal and biochemical experiments using metabolic rodent model.

**Animal Experiment:** In some projects, we use rodent models (mouse and rat) under permission from the Animal Research Ethical Committee of the TMDU. In animal studies, animals are euthanized after the experiments to collect tissue and blood samples.

Recent relevant publications from our laboratory:

1. S. Hagita, M. Osaka, K. Shimokado, M. Yoshida. Adipose inflammation initiates recruitment of leukocytes to mouse femoral artery: Role of adipovascular axis in chronic inflammation **PLoS One** 6: e19871 (2011)
2. S. Hagita, M. Osaka, K. Shimokado, M. Yoshida Combination of amlodipine and atorvastatin synergistically reduces leukocyte recruitment to mechanically injured mouse femoral artery **Hypertens Res** 34:450-455 (2011)
3. S. Ito, M. Osaka, Y. Higuchi, F. Nishijima, H. Ishii, M. Yoshida. Indoxyl sulfate induces leukocyte-endothelial interactions through upregulation of E-selectin **J. Biol. Chem**. 285:38869-38875 (2010)
4. J. Ino, C. Kojima, M. Osaka, K. Nitta, M. Yoshida. Dynamic Observation of Mechanically-injured Mouse Femoral Artery Reveals an Anti-inflammatory Effect of Renin Inhibitor. **Arterioscler. Thromb. Vasc. Biol**. 29: 1858-1863 (2009)
5. M. Nomura, H. Ishii, A. Kawakami, M. Yoshida. Inhibition of Hepatic Neiman-Pick C1-Like 1 Improves Hepatic Insulin Resistance. **Am J. Physiol.** 297: 1030-1038 (2009)
6. S. Hagita et al. Oxidative stress in mononuclear plays a dominant role in their adhesion to mouse femoral artery after injury. **Hypertension** 51: 797 (2008)
7. M. Osaka et al. Real time imaging of mechanically injured-femoral artery in mouse revealed a biphasic pattern of leukocyte accumulation. **Am. J. Physiol.** 292:H1876 (2007)

## *12 Department of Gastroenterology and Hepatology*

**Project Title:** Development of Novel Strategies for the Treatment of IBD

**Supervisor:** Prof. Mamoru Watanabe - mamoru.gast@tmd.ac.jp

**Key Words:** inflammatory bowel disease (IBD), ulcerative colitis, Crohn's disease

**Subject : Development of a novel immunoregulatory therapy for inflammatory bowel disease**

We have extensively studied the pathogenetic mechanisms of development and the persistence of chronic inflammation in the gut, aiming at identifying key factors responsible for each of these processes and devising a novel strategy for the treatment of inflammatory bowel diseases (IBD) in human. By taking integrated approaches in immunology and molecular biology, we are currently investigating the mechanistic basis for the activation of pathogenic T lymphocytes in both initial and chronic phases of gut inflammation and the mechanisms of suppressive action of CD4+CD25+ regulatory T cells in these processes. We are now focusing on several specific factors that are essential for the maintenance of pathogenic T cells as well as CD4+CD25+ regulatory T cells, so the applicants for our lab could be involved in any of these projects that they are interested in. Our research covers not only cell biology experiments or the manipulation of animal models but also the analysis of clinical samples obtained from a number of IBD patients, providing the applicants with a valuable opportunity to learn how to design and manage experiments in the field of clinical translational research.

## *13 Department of Neurology and Neurological Science*

**Project title:** "Researches on pathogenic mechanism of neurodegenerative diseases and development of their RNAi and stem cell therapies"

**Supervisor**: Prof. Hidehiro Mizusawa – h-mizusawa.nuro@tmd.ac.jp

**Key Words:** Spinocerebellar ataxia (SCA), Multiple system atrophy (MSA), Alzheimer disease, cultured cell, gene transfer, RNA interference, stem cell

**Subject #1: Study on the pathogenic mechanisms of SCA and multiple system atrophy (MSA)**

This subject aims to gain insights into degenerative disorders, particularly SCA6, an autosomal dominant cerebellar ataxia caused by polyglutamine expansion in the alpha 1A Ca channel, and multiple system atrophy (MSA), a sporadic condition showing progressive ataxia, parkinsonism and autonomic dysfunction. Variable research techniques are used: PCR, RT-PCR, laser capture microdisection, gene transfection and assays on cultured cells and fruits files, western blotting, immunohistochemistry and *in situ* hybridization on human specimens, etc *(Ishikawa K. et al. Am J Hum Genet 77:280, 2005; Watase K. et al. PNAS 105:11987, 2008; Ishiguro T. et al. Acta Neuropathol 119:447, 2010)*.

Our lab (subject #1) received one student in the year 2009. Her research will lead to open a new horizon for polyglutamine diseases.

We also study a new autosomal dominant ataxia, SCA31, caused by a non-coding pentanucleotide repeat expansion. We identified the cause of this disease after a long effort of positional cloning *(Sato N. and Amino T. et al. Am J Hum Genet 85:544, 2009)*. We have launched a new research to dissect pathogenic mechanism, using cell culture and animal models. We also utilize embryonic stem cells to widen our perspective.

**Subject #2: Targeting molecule enhancing Alzheimer phenotype caused by chronic oxidative stress**

Increased oxidative damage associated with aging is a prominent and early feature in Alzheimer disease (AD). We previously generated alpha-tocopherol transfer protein knockout (Ttpa-/-) mice, in which lipid peroxidation in the brain was significantly increased by complete depletion of a-tocopherol (a-Toc); increased thiobarbituric acid-reactive substances (TBARS) with aging *(Yokota T. et al. PNAS 98:15185, 2002)*. We crossed AD transgenic (APPsw) model mice (Tg2576) with Ttpa-/- mice. The resulting double-mutant (Ttpa-/- APPsw) mice showed earlier and more severe cognitive dysfunction in the Morris water maze, novel-object recognition, and contextual fear conditioning tests. They also showed increased amyloid beta-peptide deposits in the brain by immunohistochemical analysis, which was ameliorated with alpha-Toc supplementation. Furthermore, we made clear that this accumulation was caused by decreased clearance of amyloid beta-peptide from the brain and liver *(BBRC 350:530, 2006; J Biol Chem 284:33400, 2009)*. In these reports we provide clear evidence indicating that chronic lipid peroxidation due to alpa-Toc depletion enhances AD phenotype in

a mouse model. This crossed mouse was considered to be a model of a sporadic AD model with aging. Here, in this semester, we investigate the molecules that exacerbate the AD phenotype with increases amyloid beta-peptide from the results of mRNA expression array and micro RNA array of the Ttpa-/- and Ttpa-/- APPsw brain tissues. We can expect to find the targeting molecule in the ttherapy of AD associated with aging.

**Subject #3: Analysis of non-human primate model of amyotrophic lateral sclerosis (ALS)**

Amyotrophic lateral sclerosis (ALS) is a fatal neurodegenerative disease characterized by progressive motoneuron loss. With overexpression of wild-type TDP-43 in spinal cord of cynomolgus monkeys, last year, we show that monkeys developed progressive motor weakness and muscle atrophy with fasciculation initiated by distal hand muscles, reminiscent of ALS patients. This monkey model, but not rat model, could produce the neuropathological hallmarks of ALS, frequent cytoplasmic mislocalization of TDP-43 associated with loss of its nuclear staining. There is species difference in TDP-43 pathology, and our monkey model recapitulates ALS pathology much better than rodent model, providing a valuable tool to study pathogenesis of sporadic ALS. Excitingly, this is the first non-human primate model of ALS better recapitulated ALS features than rodent model, and its oral presentation was highlighted in the symposium of American Academy of Neurology, 2011. Using spinal cord and spinal motoneuron obtained with laser micro-dissection method from this monkey model, neuropathological, biochemical and molecular biological analysis are planned to investigate the cell death of motoneuron by overexpressed TDP-43. Furthermore, using miRNA/mRNA array analysis, we found key molecules possibly related to the pathogenesis of sporadic ALS.

**Subject #4: Gene therapy of neurodegenerative diseases using RNA interference**

RNA interference (RNAi) is a powerful tool for the post-transcriptional gene silencing. Small interfering RNA (siRNA) binds and cleaves the targeted RNA in sequence-specific manner. One possible therapeutic application of siRNA is the silencing mutant gene in autosomal-dominant neurodegenerative diseases or key molecules closely related to its pathogenesis. We had demonstrated that such an RNAi therapy is effective to cure the familial ALS using RNAi transgenic mouse *(Saito Y. et al. J Biol Chem 2005; Arch Neurol 2006)*, and developed the mutant allele-specific gene targeting strategy to escape the side effect *(Kubodera T. et al. Hum Gene Ther, 2011)*. Furthermore, we had published efficient systemic delivery of siRNA to the liver by conjugation of vitamin E *(Nishina K. et al., Mol Ther, 2008)*, and using this patented method, we recently succeeded to inhibit neuronal gene with intra-ventricular and intravenous injection of siRNA *(Uno Y. et al. Hum Gene Ther, 2011; Kuwahara H. et al, Mol Ther, 2011)*. Here, we try to make more efficient delivery system of siRNA to central nervous system to cure these neurodegenerative diseases. This project has been studied by two Imperial College students in 2008-2011.

**Subject #5: Study on modulators of amyloid-beta production**

Modulation of gamma-secretase accompanying with changing the ratio of amyloid-beta 40/42 or the amount of amyloid-beta production is one of the most important and strongest strategies

for a treatment of Alzheimer's disease. Gamma-secretase have been proved to be composed of essential four factors, presenilin, nicastrin, Aph-1 and Pen-2. We have already clarified some basic aspects of each components *(J Biol Chem 278:7374, 2003; J Biol Chem 279;46455, 2004)*. The secretase activity was reported to be regulated by a few endogenous modulation molecule and clarified its mechanism in culture cells *(Nature 440;1208, 2006)*. Some medical agents have also been reported to have the modulation effect on gamma-secretase. Effects and mechanism of new candidates will be screened and investigated using culture cells.

**Subject #6: Development of neuroreconstructive therapies for neurological disorders**
Stem cell-based therapies have a promising potential to regenerate damaged brain in neurological disorders. We focus on brain injury repair using 1) neuronal or mesenchymal stem cell transplantation (*J Neurosci Res 78:215, 2004; J Neurosci Res 88:3598, 2010*), 2) promotion of endogenous neural progenitor cell proliferation (*Proc Natl Acad Sci U S A 103:7112, 2006; Exp Neurol 207:302, 2007*), and 3) immunomodulation by means of priming regulatory T cells to promote the survival of newly-generated neurons after stroke (*J Cereb Blood Flow Metab 29:606-20, 2009*), and have found the remarkable effects of these stem cell-based therapies on neurological dysfunctions in rodent stroke models. To enhance efficacy of neuroreconstructive therapies, we are currently investigating optimal niche for stem cell mobilization by modulating inflammation and cerebral blood flow (i.e. arteriogenesis) in the stroked brain.

**Animal experiment:** Many mice and rats are used in our laboratories. No experiments using animals, however, are planned in this particular project for foreign students.

## *14 Department of Paediatrics and Developmental Biology*

**Project title:** Developmental biology in human haematology/oncology

**Supervisor:** Prof. Shuki Mizutani - smizutani.ped@tmd.ac.jp

**Key words:** leukaemia, DNA damage response, ATM, adipocyte differentiation, haematopoietic stem cells

**Study on the biological role of DNA damage response in organogenesis and oncogenesis**

ATM is the causative gene of ataxia telangiectasia, and plays a major role in protection from DNA damage and oxidative stress. We have recently uncovered a critical role of ATM in prevention of chromosomal translocation following DNA double strand breaks. Study is currently under way to disclose pathogenesis of infantile leukemia by using model mice that have a defect in DNA repair system.

Our study has also demonstrated that ATM is involved in proliferation and differentiation of stem cells and progenitor cells. *In vitro* system to track differentiation process of hematopoietic cells, hepatocytes, and adipocytes was set up in our laboratory. Our study includes ATM-dependent regulation of C/EBP family protein, a key regulator of cell differentiation, and on the impact of defective cell-cycle regulation/cell differentiation in ATM-deficiency on cancer development.

We also investigate key players in DNA damage responses, such as Artemis, Mre11/Rad50/Nbs1, and XLF, focusing on their roles in DNA repair and anti-cancer barrier.

**Animal Experiment:** Experiment on ATM-null mice is required as a part of the proposed research work (#1); however, students who do not wish to work on mice can focus on various biological & biochemical studies on primary cells and cell lines.

## *15 Department of Psychiatry and Behavioral Sciences*

**Project title:** “Single nucleotide polymorphism (SNP) analysis of mental disorders”

**Supervisor:** Prof. Toru Nishikawa - tnis.psyc@tmd.ac.jp

**Key words:** antipsychotics, dopamine, methamphetamine, NMDA receptor, phencyclidine, schizophrenia, D-serine

**Subject:** Molecular genetics of schizophrenia and mood disorders

We have been focusing on the molecular mechanisms underlying the pathophysiology of the symptoms of schizophrenia. Methamphetamine, an indirect dopamine agonist, and phencyclidine, a channel blocker of NMDA type glutamate receptor, are known to induce the psychotic symptoms such as hallucination and delusional ideas similar to schizophrenia. These drugs also induce abnormal behaviors in the rats. We have isolated a couple of genes that are responsive to these psyhotomimetic drugs from the rat cerebral neocortex. In addition, we have recently identified several molecules that may be involved in the regulation of D-serine, an endogenous NMDA receptor co-agonist in the brain. In this study, SNPs (single nucleotide polymorphisms) and VNTR (variable number of tandem repeat) of methamphetamine- and phencyclidine-responsive genes, D-serine-related genes and other genes of interest will be examined in schizophrenia and mood disorders.

**Animal experiment:** none

## *16 Department of Dermatology*

**Project title:** Functional roles of prostaglandin D2 in allergic skin diseases.

**Supervisor:** Prof. Hiroo Yokozeki **-** 3064derm@tmd.ac.jp

**Key words:** Basophils, CRTH2, DP, Eosinophils, PGD2

**Subject:** Prostaglandin D2 (PGD2) is an arachidonic acid metabolite that has long been implicated in allergic reactions. A major source of PGD2 in the skin is mast cells.
We have recently demonstrated that human dendritic cells also express hematopoietic-type PGD synthase (H-PGDS) and function as a source of PGD2. In addition, PGD2 plays an essential role in mouse models of chronic allergic inflammation. CRTH2 is preferentially expressed in Th2 cells, eosinophils, basophils, and provides stimulatory signals in those cells. Thus, PGD2 and its receptors, CRTH2 and DP, could be potent therapeutic targets for treatment of allergic skin diseases. With mice lacking *CRTH2 gene, DP gene, CRTH2/DP genes, H-PGDS gene, L-PGDS (lipocalin-type PGDS) gene, and H/L-PGDS genes,* pathophysiological roles of PGD2 and its receptors in several types of allergic skin inflammations are being studied.

**References:**

1. Satoh T, et al., 2006. Prostaglandin D2 plays an essential role in chronic allergic inflammation of the skin via CRTH2 receptor.
2. Shimura C. et al., 2010. Dendritic cells express hematopoietic prostaglandin D synthase and function as a source of prostaglandin D2 in the skin.

## *17 Department of Urology*

**Project title:** “Translational research on heat shock protein 90 (Hsp90)-targeting therapy against urologic cancer”

**Supervisor:** Prof. Kazunori KIHARA - k-kihara.uro@tmd.ac.jp
Assistant Prof. Fumitaka KOGA – chief.uro@tmd.ac.jp

**Key words:** urologic cancer, molecular targeting therapy, heat shock protein 90, treatment resistance, invasion, metastasis

**Subject #1: Can Hsp90 inhibitor overcome chemotherapy resistance of urologic cancers?**

Stability and function of many oncoproteins including mutant p53, ErbB2, Akt, survivin, and HIF-1α, are dependent on a molecular chaperone protein Hsp90. Inhibition of Hsp90 chaperoning function destabilizes these Hsp90 client oncoproteins. Hsp90 inhibitors are now progressing to phase II clinical study.

Cancer resistance against chemotherapy and radiotherapy is one of the most important clinical issues. Some of the Hsp90 client oncoproteins are associated with resistance of cancer cells against chemotherapy and radiotherapy. Our hypothesis is that Hsp90 inhibitors would overcome the treatment resistance of cancer by suppressing Hsp90 client oncoproteins associated with anti-apoptotic signaling. We are studying sensitizing effects of Hsp90 inhibitors on human urologic cancer cells to chemotherapeutic agents commonly used in the clinic, such as cisplatin, gemcitabine, and docetaxel.

**Subject #2: Suppression of cancer invasion and metastasis by Hsp90 inhibitor**

Cancer is life-threatening disease because of its invasive and metastatic potential. Hsp90 inhibitors could suppress multiple signaling pathways involved in invasion and angiogenesis. Our hypothesis is that low-dose Hsp90 inhibitor would keep tumor dormancy and prolong survival of cancer-bearing mice by suppressing cancer invasion and metastasis.

**Animal experiment:** We plan *in vivo* experiments using cancer-bearing nude mice model. In subject #1, mice model of metastatic cancer is made by injecting cancer cells into the tail vein or into the left ventricle. In subject #2, cancer cells of highly metastatic potential are inoculated subcutaneously. Mice bearing cancer are sacrificed at appropriate time points to evaluate therapeutic effects of Hsp90 inhibitor.

## *18 Department of Otolaryngology*

**Project Title:** Genetic analysis of the deafness genes in families with autosomal dominant hereditary hearing loss

**Supervisor:** Prof. Ken Kitamura - kitamura.oto@tmd.ac.jp

**Key words:** Hereditary Hearing Loss, Deafness Gene, Identification of Mutations

**Subject:** Genetic analysis has been applied to improve understanding of non-syndromic hereditary hearing loss and is useful for genetic counseling purposes. However, the identifying rate of a causative gene mutation is not necessarily high. In order to increase the rate of the identification of the deafness gene mutations, we focused a founder effect of the deafness genes. We recently screened for the two mutations in *DFNA5* in a total of 96 unrelated Japanese patients with autosomal dominant non-syndromic hereditary hearing loss and we demonstrated that two (2%) of the 96 patients carried the mutations in *DFNA5*. These two families were the sixth and the seventh family in which mutations in *DFNA5* was identified in the world.
In this subject the materials are the DNA which have been extracted from peripheral blood lymphocytes using standard methods after obtaining written informed consent from the 96 patients with autosomal dominant non-syndromic hereditary hearing loss. We target the deafness genes whose mutations were identified in East Asians. The relevant segment of DNA of the candidate deafness genes is amplified by polymerase chain reaction (PCR). The PCR products are purified and directly sequenced to identify the mutations. Whenever the mutation is detected, amplification and sequencing are conducted for all coding regions and exon-intron boundaries of the candidate genes. Animal experiments are not included in the subject.

**References:**

1. Abe S, Noguchi Y, Kitamura K: What do patients with hereditary deafness think of genetic studies? Auris Nasus Larynx 37: 422-6, 2010.
2. Yashima T, Noguchi Y, Kawashima Y, Rai T, Ito T, Kitamura K: Novel ATP6V1B1 mutations in distal renal tubular acidosis and hearing loss. Acta Otolaryngol 130: 1002-8, 2010.
3. Fujikawa T, Noguchi Y, Ito T, Takahashi M, Kitamura K: Additional heterozygous 2507A>C mutation of *WFS1* in progressive hearing loss at lower frequencies. Laryngoscope 120: 166-71, 2010.
4. Noguchi Y, Ito T, Nishio A, Honda K, Kitamura K. Audiovestibular findings in a branchio-oto syndrome patient with a SIX1 mutation. Acta Otolaryngol. 131:413-8, 2011

## *19 Department of Metals*

**Project title:** Biofinctionalization of metals

**Supervisor:** Prof. Takao Hanawa – hanawa.met@tmd.ac.jp

**Keywords:** biomaterial, metal, alloy, biocompatibility, biofunction, functional molecule, surface technology, surface anlysis, cell culture

**Subject #1: Inhibition of formation of biofilm on metals**

Biofilm formation inducing infectious desease is a major cause for retrieval of medical implants in orthopedics. To prevent the formation of biofilm, we attempt the following approaches. We could control bonding manner of poly(ethylene glycol), PEG, to metal surfaces with electrodeposition at present and experiments to evaluate biofilm formation are designed on trial and error with collaborating a laboratory in the dental school. We have investigated factors influencing bonding strength and durability at the interface between metals and polymer. In the next stage, we attempt surface modification of polymer with ion beam and immobilization of functional radicals to inhibit the formation of biofilm.

**Subject #2: Prevention of artifact under MRI**

Metals show a low magnetic susceptibility or antimagnetic materials are required for implant devices form the viewpoint of the imaging of MRI. In addition, these materials are required for medical devices and instruments for operations and treatments used under open MRI. We are attempting the following approaches. We have found that the addition of Nb decrease magnetic susceptibility of Zr which shows originally low magnetic susceptibility and that the metallurgical structure governs magnetic susceptibility. Therefore, we must investigate more detail about the effect of alloying elements and structure.

**Subject #3: Control of hard tissue compatibility**

Both high bone conductivity and inhibition of bone formation are required according to implants' purpose. Therefore, we attempt to add both properties to metals with the following techniques. We have found that more RGD is immobilized on titanium through electrodeposited PEG than without PEG and calcification on the RGD/PEG/Ti material is much larger than those on Ti and RGD/Ti.. Therefore, we deeply investigate this phenomenon to improve hard tissue compatibility of Ti alloys. Zr does not form calcium phosphate on itself that is a excellent property for intraosseous bone fixators. Here, we attempt the bone formation on Zr to develop materials having both high bone conductivity and inhibition of bone formation according to the parts. With cathodic polarization, we could obtain alkaline near the surface and calcium phosphate easily formed there. We have to examine the condition of the polarization.

**Subject #4: Soft tissue adhesion**

Soft tissue adhesion is necessary in dental implants, housing of pacemakers, devices passing through skin, and so on. No materials adhesive to soft tissue has been developed. In this subject, we will approach to the goal with the following three paths. Immobilization of adhesive factors on metals through electrodeposited PEG will be attempted and soft tissue adhesion to factor/PEG/metal surface must be evaluated. We are attempting the unidirectional immobilization of collagen to metal surfaces by electrodeposition without any denaturalization, and have partly succeeded it at present. We investigate conditions for electrodeposition such as pH, temperature, voltage, and so on.

## *20 Department of Biomedical Information (Biosystems)*

**Project Title:** On-chip Cellomics Technology Developlemt for Predictive *in-vitro* Drug Discovery

**Supervisor:** Prof. Kenji YASUDA - yasuda.bmi@tmd.ac.jp

**Key words:** hES cells, hiPS cells, cardiomyocyte cells, neuronal cells, on-chip screening, MEMS, nanobiomedicine, drug discovery, toxicology

**Subject #1:** Studies on Epigenetic Information Stored in Living System

In this subject, we have examined a series of studies to analyze emergence of order in the spatiotemporal structures of cell network to expand our understanding of how the emergence of the order in living systems is determined. As cells are minimum units reflecting epigenetic information, which is considered to map the history of a parallel-processing recurrent network of biochemical reactions, their behaviors cannot be explained by considering only conventional simple one-way 'self-organization' process regulated by DNA information, especially during the cell division process. The role of emergence of order in the higher complexity of celluler groups, which complements their genetic information, is inferred by comparing predictions from genetic information with cell behaviour observed under conditions chosen to reveal adaptation processes and community effects. A system for analyzing emergence of order will be developed starting from the twin complementary viewpoints of cell regulation as an 'algebraic' system (emphasis on temporal aspects; adaptation among generation) and as a 'geometric' system (emphasis on spatial aspects; spatial pattern-dependent community effect). The acquired knowlege may lead not only to understand the mechanism of the inheretable epigenetic meory but also to be able to control the epigenetic information by the designed sequence of the external stiulation. In practice, students will measure the epigenetic information in living systems such as brain (neuronal network system), immune system.

**Subject #2:** Constructing “On-chip Quasi-*in vivo* Model” using Nano-Bio Technology

Using constructive approach, we are developing artificial organ model on chip for drug discovery and toxicology use. Especially, using hES/hiPS cell-delivered cardiomyocyte cells, we are developing the preclinical cardiotoxicology screening system on a chip. This system has an potential to measure the risk of TdP occurrence.

**Animal Experiment:** Students are expected to learn how to culture hES/hiPS cell-delivered cells on the biochips. Animal experiments are not planned in these subjects.

## *21 Department of Biomedical Devices and Instrumentation*

**Project title:** Advanced Biomedical Sensors and Bioinstrumentation.

**Supervisor:** Prof. Kohji MITSUBAYASHI - m.bdi@tmd.ac.jp

**Key words:** Human-MEMS sensors, Non-invasive monitoring, Bioelectronic-sniffers, Volatile biomarker analysis

**Subject #1:** Wearable chemical sensors for non-invasive bio-monitoring

We have developed flexible and wearable chemical sensors for non-invasive bio-monitoring. This sort of sensors is fabricated using Soft-MEMS (Micro Electro Mechanical System) technology for functional polymers. We are also working on advanced bio-monitoring using flexible biosensor for continuous in-vivo monitoring of glucose in tear fluids. The change of tear glucose level induced by oral-administration of glucose was successfully monitored by animal experiment in our previous study. We are currently studying non-invasive approaches of transcutaneous gas monitoring using the flexible chemical sensors.

**Subject #2:** Biochemical gas sensors for volatile biomarker analysis

High-selective biochemical gas sensors (Bio-sniffers) and its application for continuous monitoring of volatile information for human bodies have been constructed with biological recognition materials such as drug-metabolizing enzyme in human liver. Previously, we demonstrated potential applications of the bio-sniffer such as objective halitosis diagnosis, high-selective aldehyde and breath alcohol monitoring after alcohol intake. We have started to develop a biological sensor for methyl mercaptan which is causative agent of the fish-odor syndrome.

**Animal Experiments:** Concerning Subject 1, animal experiments are planned using Japan white rabbit. Animal experiments are not planned in Subject 2.

## *22 Department of Bioelectronics*

**Project Title:** Sensing methodology for bio-molecular recognition and cell functions

**Supervisor:** Prof. Yuji Miyahara - miyahara.bsr@tmd.ac.jp

**Key words:** bio-transistors, point of care testing, biomarkers, surface modification

**Subject 1: Electrical Detection of Circulating MicroRNAs for Cancer Diagnosis**

We have been investigating direct interaction between biomolecules and a solid-state substrate. We proposed novel concept of biologically coupled field effect transistors (FET) which is based on direct transduction of charge density change of biomolecules into electrical signal by the field effect. We are currently investigating a new method to detect circulating microRNAs simply using the bio-FET, since microRNAs may serve as a diagnostic marker for cancer.

**Subject 2: Non-destructive Monitoring of Transporter-substrate Interaction at Cell Membrane**

We investigate a cell-based field effect transistor (cell-based FET) for drug transport analysis, in which target transporters are expressed at the cell membrane. Non-destructive and real-time monitoring of the uptake kinetics of substrates mediated by membrane-bound transporters can be realized with oocyte-based FET. Discrimination of transporting ability among genotypes of the transporters can be achieved just by placing the oocyte on the gate surface. The platform based on the cell-based FETs is suitable for high-throughput screening in pharmaceutical lead discovery.

**Animal Experiments: None**

**References:**

1. A. Matsumoto, H. Cabral, N. Sato, K. Kataoka, Y. Miyahara, Assessment of Tumor Metastasis via Direct Determination of Cell Membrane Sialic Acid Expression, Angew. Chem. Int. Ed., 49, 5494-5497 (2010)
2. A. Matsumoto, N. Sato, H. Cabral, K. Kataoka, and Y. Miyahara, Noninvasive Sialic Acid Detection at Cell Membrane by Using Phenylboronic Acid Modified Self-Assembled Monolayer Gold Electrode, *J. Am. Chem. Soc*., 131, 12022–12023 (2009)
3. T. Sakata and Y. Miyahara, Noninvasive monitoring of transporter-substrate interaction at cell membrane, Anal. Chem. 80, 1493-1496 (2008)
4. T. Sakata and Y. Miyahara, DNA sequencing based on intrinsic molecular charges, Angew. Chem. Int. Ed., 45, 2225-2228 (2006)

## *23 Department of Molecular Medicine and Metabolism*

**Project title:** Fighting obesity and the metabolic syndrome!

**Supervisor:** Prof. Yoshihiro Ogawa - ogawa.mmm@mri.tmd.ac.jp

**Key words:** adipocytes, cytokines, diabetes, macrophages, metabolic syndrome, obesity, skeletal muscle, transcription factors

The mission of our laboratory is to elucidate the molecular mechanism underlying obesity and the metabolic syndrome, thereby contributing to the improvement of our “quality of life”.

**Subject #1: Molecular mechanism of adipose tissue remodeling**
Obesity is a state of chronic, low-grade inflammation. Obese adipose tissue is characterized by adipocyte hypertrophy, followed by increases in angiogenesis, macrophage infiltration, and unbalanced production of pro-inflammatory vs. anti-inflammatory adipocytokines, suggesting the previously unrecognized dynamic changes, which may be referred to as “adipose tissue remodeling”. In **Subject #1**, we pursue the molecular mechanism underlying adipose tissue remodeling and thus identify novel therapeutic targets that may reduce obesity-related inflammation and the metabolic syndrome.

**Subject #2: Transcriptional regulation of skeletal muscle metabolism**
A well-balanced body energy budget controlled by limitation of calorie uptake and/or increment of energy expenditure that is typically achieved by proper physical exercise is effective against obesity and diabetes. The skeletal muscle is the largest organ in the body and plays an important role in exercise, energy expenditure, and glucose metabolism. The mass and glucose and lipid metabolism of the skeletal muscle is regulated in response to changes in physical activity, environment, or pathologic conditions. In **Subject #2**, through the cell- and animal-based approaches, we pursue the transcriptional regulation of skeletal muscle metabolism and thus identify new therapeutic strategies against skeletal muscle atrophy and abnormal glucose and lipid metabolism.

**Animal Experiments:** We use transgenic and knockout mouse models of obesity and the metabolic syndrome for *in vivo* experiments. The exchange student may or may not join the animal experiments.

## *24 Department of Molecular Pharmacology*

**Project title:** Molecular regulation of calcium metabolism

**Supervisor:** Prof. Masaki Noda - noda.mph@mri.tmd.ac.jp

**Key words:** calcium, osteoblasts, nervous system bone.

**Background:**
In order to contribute to the establishment of therapeutic and preventive methods for osteoporosis and other calcium-balance related disorders, we are elucidating molecular mechanisms underlying regulation of bone formation and resorption, with emphases on calcium homeostasis. Skeletal system is a largest storage site for calcium in a living-body and its homeostatic balance is achieved through complex cellular network consisting of bone-forming osteoblasts, bone-resorbing osteoclasts, bone marrow stromal cells and osteocytes, as well as by network from endocrine systems. To investigate the mechanisms of regulation in this balance during development and differentiation, we use molecular, genetic and cell biological methods focusing on genes of our interests.

**Subject:**
**Constitutively active parathyroid hormone receptor signaling in cells in osteoblastic lineage will be** investigated to explain the roles of parathyroid hormone receptor (PTH/PTHrP receptor) signaling in osteoblasts. We will transfect plasmid enconding active receptor. This would increase cell proliferation and differentiation. Signaling by constitutively active PTH/PTHrP receptor (caPPR), will regulate the osteoblast-specific Col1a1 promoter (Col1a1-caPPR), would change cell behavior. In Col1a1-caPPR transfected cells it will increase expression of genes encoding bone formation markers. We will examine the expression of markers and mineralized nodule formation in vitro. Project will be to test PTH-induced enhancement of bone formation parameters.

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## *25* Department of Stem Cell Regulation

**Project title:** Molecular regulation of mouse neural stem cells, mouse fetal hematopoietic stem cells, and rat glioma stem cells

**Supervisor:** Prof. Tetsuya Taga - taga.scr@mri.tmd.ac.jp
Assoc. Prof. Tetsushi Kagawa - kagawa.scr@mri.tmd.ac.jp
Assoc. Prof. Ikuo Nobuhisa - nobuhisa.scr@mri.tmd.ac.jp

**Key words:** neural stem cell, stem cell fate, brain development, epigenetic regulation of neural development, hematopoietic stem cell, fetal hematopoiesis, cancer stem cell, stem cell niche

**Subjects:**

During the exchange program, the course in this department will introduce to students the recent topics in the research field of stem cell regulation. Tissue stem cells possess potential to generate all cell types present in a given tissue. In order to understand tissue development and regeneration from the biological and clinical viewpoints, it is important to study the molecular regulation of stem cell maintenance and fate specification.

Not only normal tissue stem cells, e.g. neural and hematopoietic stem cells that we place particular focus on, but also cancer stem cells will be discussed to consider the problem of cancer recurrence. Particular attention is given to cell-external cues (such as cytokines) and cell-intrinsic programs (including chromatin modification), taking cross-interactions of transcriptional regulatory signals into consideration.

Students will receive exposure to concepts and research technologies related to these research areas, and conduct a research project under supervision of the supervisors, on regulatory mechanisms in either (1) neural stem cells in the developing mouse brain, (2) hematopoietic stem cells in hematopoietic tissues in the mouse fetus, and (3) cancer stem cells in rat glioma cells. Students are advised to design experiments regarding, for example, stem cell development, maintenance of multipotentiality, cell-fate specification, cell migration, maturation, maintenance, and regeneration. Through execution of such experiments, students shall understand general property of stem cells in both/either physiological and/or pathological conditions and obtain a hint for going into translational research. The project options planned for students during the course are as follows:

1. Signals regulating maintenance and cell-fate determination of neural stem cells
2. Epigenetic regulation of central nervous system development and function
3. Regulation of hematopoietic stem cells during fetal development
4. Molecular signature of cancer stem cells and their niche

## *26 Department of Neuropathology*

**Project title:** Genotoxic Stress and Neurodegeneration

**Supervisor:** Prof. Hitoshi Okazawa - okazawa.npat@mri.tmd.ac.jp

**Background:**
Neurodegenerative diseases are caused by aggregation of misfiled disease proteins inside or outside of neurons. The disease proteins are suspected to interact with physiological cellular proteins before aggregation, and to dysfunction or decrease the normal proteins. In polyglutamine diseases including 9 neurodegenerative diseases such as Huntington's disease and familial spinocerebellar ataxias, it is known that nuclear translocation of the disease proteins is essential for the pathology. We have shown functionally change of several molecules involved in DNA damage repair contributes to the pathology by using cellular, fly and mouse models.

Hoshino M et al. (2006) Transcriptional repression induces a slowly progressive atypical neuronal death associated with changes of YAP isoforms and p73. ***Journal of Cell Biology*** 172, 589-604.

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Enokido Y et al. (2010) Mutant huntingtin impairs Ku70-mediated DNA repair. ***Journal of Cell Biology*** 189, 425-443.

**Project**
1) To clarify the molecular connection between nuclear and cytoplasmic pathologies in neurodegeneration.
2) To find other genes affecting neurodegeneration.

## *27 Department of Molecular Pathogenesis (in Medical Research Institute), Laboratory of Genome Diversity (in School of Biomedical Science)*

**Project title**：Structure and function of human genome diversities involved in the pathogenesis of heart failure, atherosclerosis, or HIV/AIDS

**Supervisor**: Prof. Akinori Kimura - akitis@mri.tmd.ac.jp

**Key words**: Human genome, Mutation, Cardiovascular disease, HIV/AIDS

**Subject #1**: Molecular pathogenesis of cardiomyopathy and heart failure
Primary cardiomyopathy is a disease condition caused by functional abnormalities of cardiac muscle resulting in heart failure. There are two major clinical types of primary cardiomyopathy; hypertrophic cardiomyopathy (HCM) characterized by cardiac hypertrophy and diastolic dysfunction and dilated cardiomyopathy (DCM) associated with dilated heart chamber and systolic dysfunction. Since 50-70% of HCM cases and 20-35% of DCM cases have apparent family history of the disease, primary cardiomyopathy can be caused by gene mutations. We have identified various gene mutations causative for HCM or DCM. However, it is still unclear how these mutations cause the disease and how we can prevent the disease. In this project, functional alterations caused by the disease-causing mutations will be investigated to find-out a strategy for prevention of cardiomyopathies.

**Subject #2**: Genes involved in the susceptibility to myocardial infarction and/or cardiovascular atherosclerosis
Myocardial infarction (MI) is a typical multifactorial disease in which genetic factors and environmental factors are involved. By using a whole-genome microsatellite association study, we have identified six different loci associated with the susceptibility to MI. Among these 6 loci, we have identified SNPs responsible for the association in 2 loci. In this project, functional implications of these disease-associated SNPs will be investigated to understand the etiological role of these MI loci.

**Subject #3**: Human genome diversities involved in the susceptibility to HIV infection and/or AIDS development
HIV infection and AIDS are one of the most severe infectious diseases in human. It is well known that not all of the individuals exposed to HIV would be infected HIV and not all of the individuals infected by HIV would develop AIDS, suggesting the presence of genetic factors controlling the HIV infection and/or AIDS development. Several gene polymorphisms have been reported to be involved in this genetic control, including polymorphisms in CCR5, CCL3L1 and HLA-B in Caucasians. However, most of the findings in Caucasians are not applicable to Japanese. In this project, polymorphisms in candidate genes will be investigated to reveal the susceptibility to HIV/AIDS in Japanese.

**Animal Experiment**: Model animals of cardiomyopathy (transgenic mice and knock-in mice) will be analyzed for the pathological features. Mice will be sacrificed following the guidelines for the Care and Use of Laboratory Animals published by the National Institute of Health (NIH Publication 85-23, revised 1985)

## *28 Department of Molecular Genetics*

**Project title:** Molecular mechanism for breast carcinogenesis

**Supervisor:** Prof. Yoshio MIKI – miki.mgen@mri.tmd.ac.jp

**Key Words:** Breast cancer, DNA damage repair, apoptosis, BRCA1, BRCA2

**Background:**

The failure of DNA damage repair function causes many kinds of disease including cancer. Especially, the breast cancer occurs due to mutations of BRCA1 and BRCA2 genes related to double stranded break repair mechanism, and the explication of the signal transduction pathway by these two molecules is indispensable to clarify the mechanism of breast carcinogenesis. To clarify the mechanism of breast carcinogenesis, we are analyzing the DNA damage repair and cell death induction.

**Subject #1: Investigation of a molecular mechanism for breast carcinogenesis**

The breast cancer susceptibility protein, BRCA2, preserves chromosomal stability through roles in the repair of DNA double-strand breaks and cell division. We have previously reported that BRCA2 may regulate the positioning of the centrosome. However, molecular mechanisms of BRCA2-based functions in centrosomes are not fully understood. Here we analyzed BRCA2 co-sedimented proteins from centrosomes of HeLa cells by mass spectrometry. We will analyze the function of the gene responsible for hereditary breast cancer, BRCA2 and its related genes.

**Subject#2: The intracellular signaling transduction and cell death in DNA damage**

- Analysis of the cell cycle control mechanism by protein kinase C delta
- The investigation of the mechanism of apoptosis induction by c-Abl

**Animal experiment: None**

## *29 Department of Molecular Epidemiology*

**Project title:** Gene-environment interaction in common diseases

**Supervisor:** Prof. Masaaki Muramatsu – muramatsu.epi@mri.tmd.ac.jp

**Key words:** diabetes, hypertension, hyperlipidemia, obesity, metabolic syndrome, atherosclerosis, human genome, single nucleotide polymorphism (SNP), association study

**Subject #1.** Gene-environmental interaction in metabolic syndrome
Metabolic syndrome is a typical multifactorial disease characterized by diabetes, hypertension, hyperlipidemia, and obesity, The etiology involves both genetic and environmental factors, but the detail is still unknown. Systemic and low grade inflammation and following insulin resistance are known to be instrumental for the pathophisiology. The aim of this project is decipher genes and gene-environment interactions that lead to the development of metabolic syndrome on the bases of a cohort study in Japan. Genotyping of SNPs in the human genome as well as biostatistical analysis will be employed in this project.

**Subject#2** Genetic factors in atherosclerosis development
Atherosclerosis is a silent disease, whose progression is not usually recognized until the outbreak of cardiovascular or celebrovascular events. The development of atherosclerosis involves both environmental and genetic factors, where the latter is estimated to contribute to 30%–50% of the risks of atherosclerosis. The aim of this project is to decipher genetic factors that risks and protects systemic atherosclerosis. More than 1,500 consecutive autopsy cases, which are evaluated with pathological atherosclerosis is employed for the study. Candidate genes are selected from the recent genome wide association studies. Involvement of epigenetic changes such as DNA methylation will also be in the scope of the research.

**Animal experiment:** none

## *30 Department of Bioinformatics and Computational biology*

**Project title:** Systems life Science

**Supervisor:** prof. Hiroshi Tanaka tanaka@bioinfo.tmd.ac.jp

**Key words:** Personalized Medicine ,Omics Based Medicine, Clinical Omics ,Systems evolutionary biology, Hox clusters

**Subject #1:** Development of Integrated Database of OMICS data and Clinical Information.

Development of “TMDU Clinical Omics Database” have been conducted to integrate both multiple comprehensive molecular biological information and comprehensive clinical information. The goal is to make an integrated database for clinical research and to analyze the mechanism of the disease.

We are currently collecting Hepatic, Colon, and Oral Cancer samples just after biopsy or surgery. Clinical Research Coordinator (CRC) gets the comprehensive clinical record and perform interview to collect the lifestyle, medical history, etc. Laser Capture Micro-dissection (LCM) is performed when needed and DNA /RNA are extracted for transcriptome and copy number variation analysis.

By analyzing the clinical phenotype and biological phenotype, new correlation has been found from the database, which currently has information from more than 200 samples. We believe this database can be the basis for realizing "Personalized Medicine" to the phase of "Translational Research" from the standpoint of “Omics Based Medicine”.

Recently, we have developed a new discipline named “systems pathology”. Most of diseases is not occurred by aberrations of one or two genes but should be considered to be occurred by distortion of molecular pathway. Systems pathology is now clarifying the invasion-metastasis of cancer from the systems level understanding of epithelial-mesenchymal transition.

**Subject #2:** Systems evolutionary biology: study on systems evolution and systems dynamics

To understand the evolution and dynamics of life as a unified system, we propose a new scientific discipline called “systems evolutionary biology”, which consider the evolution as complexification process of molecular network. We have analyzed systems evolution of *Hox* signaling pathway from the systems evolutionary biology. *Hox* genes form regulatory systems of an early development in a metazoan embryo. We have shown that the *Hox* clusters achieved higher-level control of the body plan by incorporating a new degree of freedom compared to its previous regulation capability through the gain/diversification of *Hox* genes and sequence motifs. It might be said that biological systems evolve hierarchically with nested structure, which means that by continuing to utilize the previous organization but gaining a new dimension, they achieve a qualitatively higher level of biological complexity. On the other

hand, we also have analyzed network structure transition during the development of iPS cell and HIV within-host evolution based on time-series microarray data by developing new methods of systems evolutionary biology.

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## *31 Department of Molecular Neuroscience*

**Project title:** Molecular pathogenesis of major mental illnesses.

**Supervisor:** Prof. Kohichi Tanaka – tanaka.aud@mri.tmd.ac.jp

**Key words:** glutamate, transporter, conditional knockout mouse, schizophrenia, autism, depression, obsessive-compulsive disorder

**Subject #1:** Molecular mechanisms of obsessive-compulsive disorder
Obsessive-compulsive disorder (OCD) is a common psychiatric disorder characterized by persistent intrusive thought and repetitive actions. In recent years, several clinical studies suggest that impairment of glutamate neurotransmission is involved in the pathogenesis of OCD. In subject #1, we clarify relationship between glutamate neurotransmission dysfunction and OCD.

**Subject #2:** Molecular mechanisms of depression
The molecular mechanisms of depressive disorders are poorly understood. Increased activity of the habenula has been implicated in the etiology of major depressive disorder. In subject #2, we clarify relationship between in creased neuronal activity of the habenula and depression.

**Animal Experiments**: We use genetically engineered mouse model of major mental illnesses. The exchange student may join the animal experiments.

## *32 Department of Immunology*

**Project title**: Molecular pathogenesis of autoimmune diseases.

**Supervisor:** Prof. Takeshi Tsubata - tsubata.imm@mri.tmd.ac.jp

**Key words:** autoimmune disease, lymphocyte, autoantibody, cell signaling, chemical compounds.

**Subject #1:** Searching for chemical compounds that regulate autoimmune diseases.

We have demonstrated that some of the B lymphocyte (B cell) membrane molecules such as CD22 and CD72 regulate development of autoimmune diseases including lupus and diabetes. By screening library of chemical compounds in the Chemical Biology Screening Center, this project aims at isolating chemical compounds that regulates moclecules such as CD22 and CD72, and examining their ability to modulate clinical course of autoimmune diseases in mouse models.

**Subject #2:** Studies on tolerance of self-reactive B cells.

In healthy individuals, self-reactive B lymphocytes that potentially produce autoantibodies are either eliminated or functionally inactivated thereby inhibiting autoantibody production. By using transgenic mice producing autoantibodies, we established a mouse model in which the fate of self-reactive B cells can be easily examined. How self-tolerance is maintained in normal individuals, and how it is ablated in mouse model of autoimmune diseases will be addressed.

**Animal experiment:** Mice are used for the study of both Subjects #1 and #2. Mice are handled according to the regulation of the university.

## *33 Department of Gene Expression*

**Project title:** Visualization of alternative pre-mRNA splicing patterns *in vivo*.

**Supervisor:** Associate Professor Hidehito KUROYANAGI kuroyana.end@tmd.ac.jp

Lab URL: http://www.tmd.ac.jp/english/end

**Keywords:** mRNA, alternative splicing, *in vivo* imaging, *C. elegans*, fluorescence reporter, transgenic animal, mutant screening

**Background:**

Alternative splicing of precursor mRNA (pre-mRNA) is an important mechanism for producing proteome diversity in multicellular organisms. Recent high-throughput sequencing analysis of human tissue transcriptomes revealed that more than 90% of human genes undergo alternative splicing and that most of these alternative splicing events vary between tissues.

The mechanisms involved in regulating alternative splicing in living cells have, in the past, been studied using splicing-reporter minigenes; alternative mRNA isoforms derived from reporter minigenes were analyzed by quantifying reverse transcription PCR (RT-PCR) products. However, the laborious nature of these procedures prevented high-throughput analysis of alternative splicing regulation in living cells or organisms.

We have recently utilized *C. elegans* as a model organism to study alternative splicing regulation *in vivo*. By constructing multi-chromatic alternative splicing reporter mini-genes, we successfully visualized cell-type-specific and developmentally regulated alternative splicing events *in vivo*. As *C. elegans* is transparent, it is easy to observe expression patterns of multiple fluorescent proteins in living worms at a single cell resolution. Furthermore, a variety of genetic tools for *C. elegans* research, such as transgenic expression of exogenous proteins, mutant screening and gene mapping, and RNAi-mediated gene knock-down, facilitated further analyses of *trans*-acting factors and *cis*-elements, and identification of partially spliced RNA species. Another advantage of *C. elegans* in studying splicing regulation is that its introns are on average very short and therefore it is easy to construct reporter mini-genes that include all the required elements.

**Subject:**

The prospective students will construct fluorescence reporter mini-genes to visualize alternative splicing patterns of genes of their interest, and generate transgenic reporter worms by microinjecting the minigenes. They will integrate the transgenes and screen for mutants defective in the tissue-specific and/or developmental regulation.

**Reference**

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**Animal Experiments:** Nematode worm, *Caenorhabditis elegans*

## *34 Department of Signal Gene Regulation (Human Gene Sciences Center)*

**Project Title:** Transcriptional regulation of cellular proliferation, differentiation and transformation

**Supervisor:** Prof. Masataka Nakamura－naka.gene@tmd.ac.jp

**Key words:** TWIST, repression, mesenchymal cell, HTLV-I, Tax, transactivation, leukemia, telomerase, cellular senescence.

**Subject #1:** Roles of TWIST in cell growth and differentiation

TWIST is a member of the basic helix-loop-helix (bHLH) transcription factor family and plays key roles in the epithelial-mesenchymal transition (EMT) and mesenchymal differentiation. It has been recently suggested a link between that tumor metastasis and EMT. We previously found that TWIST keeps mesenchymal progenitor cells growing through repression of transcription of the $p21^{WAF1/Cip1}$gene, raising questions of how TWIST represses transcription in mesenchymal cells and of how TWIST expression is controlled during EMT and mesenchymal differentiation. We are currently addressing these issues by means of genetic and molecular approaches.

**Subject #2:** Molecular basis of leukemogenesis by human T-cell leukemia virus type I.

Tax is a transcriptional protein of the human retrovirus HTLV-I, which is a causative agent of adult T-cell leukemia (ATL). In general, cellular transformation requires three major alternations in normal cell behavior; abnormal proliferation, apoptosis prevention and immortalization. We and others have extensively studied molecular basis of Tax-mediated cell growth and prevention of apoptosis. We have recently demonstrated Tax-dependent modulation of telomerase (hTERT) gene expression that is critical for cellular immortalization, and identified two hTERT promoter elements responsible for hTERT expression. Function of the nuclear factors, which bind to the elements, is currently investigated.

**No animal experiment is planned**.

### *35 Department of Surgical Oncology*

**Supervisor:** Professor Kenichi Sugihara

**Contact Information;**

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Department of Translational Oncology,

Research Subjects

- Nerve-preserving surgery for colorectal cancer
- Staging of liver metastasis from colorectal cancer
- Neoadjuvant and conversion chemotherapy for liver metastasis from colorectal cancer.
- Laparoscopic surgery for gastrointestinal cancer with lymphnode dissection
- Diagnostic laparoscopy and intraperitoneal administration of anti-cancer agent for far advanved gastric cancer
- Nerve-preserving surgery for gastric cancer
- Tailor-made therapy for breast cancer, including primary reconstruction after resection of the breast
- Molecular detection of predictive markers in chemotherapy for gastrointestinal and breast cancer
- Clinical trials of chemotherapy for gastrointestinal cancer - efficacy of combination chemotherapy and molecular targeted agents -