Center of Excellence (COE) Program

Project of Mesenchymal Stem Cells
研究拠点形成　間葉系幹細胞プロジェクト

Project Associate Professor　Hideaki Nakajima, M.D., D.M.Sc.　特任准教授　医学博士　中　島　秀　明

Our major goal is to dissect regulation of hematopoiesis-self-renewal of hematopoietic stem cells (HSC) and the regulation of hematopoietic cell differentiation by transcription factors, and translate this knowledge into clinical settings to provide novel therapies for patients with hematological diseases.

1. Immune suppressor factor confers bone marrow stromal cells with enhanced supporting potential for hematopoietic stem cells

Hideaki Nakajima, Fumi Shibata[1], Yumi Fukuchi[1], Yuko Goto-Koshino[1], Miyuki Ito, Hiroyuki Aburatani[2], Toshio Kitamura[1]: [1]Division of Cellular Therapy, Advanced Clinical Research Center, The Institute of Medical Science, The University of Tokyo [2]Department of Cancer Systems Biology, Research Center for Advanced Science and Technology, The University of Tokyo

Immune suppressor factor (ISF) is a subunit of the vacuolar ATPase proton pump. We earlier identified a short form of ISF (ShIF) as a stroma-derived factor that supports IL-3-independent growth of a mutant subline of Ba/F3 cells. Here we report that ISF/ShIF not only supports a mutant Ba/F3, but also self-renewal and expansion of primary hematopoietic stem cells (HSC). Co-culture of murine bone marrow cells with a stromal cell line overexpressing ISF or ShIF (MS 10/ISF or MS10/ShIF) significantly enhanced their colony-forming activity and the numbers of long-term culture initiating cells (LTC-IC). Moreover, competitive repopulating activity of c-Kit＋Sca-1＋Lin- HSC was significantly maintained by co-cultivation with MS10/ISF or MS 10/ShIF. These stem cell supporting activities were abolished in the proton pump mutant of ISF/ShIF, indicating that proton transfer across cellular or endosomal membrane was critical. Gene expression analysis of ISF/ShIF-transfected cell lines revealed downregulation of secreted frizzled related protein (SFRP)-1 and tissue inhibitor of metalloproteinase-3 (TIMP-3), and the restoration of SFRP-1 and TIMP-3 expressions in MS10/ISF cells partially reversed its enhanced LTC-IC supporting activity to a normal level. These results suggest that ISF/ShIF supports HSC by modulating Wnt-activity and the extracellular matrix, and provide new insights in HSC physiology *in vivo*.

2. Molecular mechanism of stem cell self-renewal on bone marrow stroma

Hideaki Nakajima, Fumi Shibata[1], Yumi Fukuchi[1] and Toshio Kitamura[1]

Hematopoietic stem cells (HSC) keep self-renewing in the bone marrow in order to support continuous blood cell production. These processes are thought to occur in the bone marrow niche, a special microenvironment created

by stromal cells. HSC-stromal cell interaction is thought to provide unknown signals to keep HSC in immature state and makes them undergo extensive self-renewal. However, molecular mechanism of these processes is poorly understood. We are trying to address this question by following approaches. 1) Identify cell surface molecules that are expressed on hematopoietic stem cells and bone marrow stromal cells by utilizing a variety of technologies (i.e. signal sequence trap, mRNA subtraction) and analyze their function in vitro and in vivo. 2) Identify secreted proteins (i.e. cytokine, growth factor, extracellular matrix protein) from the bone marrow stroma that are potentially important for HSC regulation. From the first approach, we identified novel HSC marker, Robo4 that is specifically expressed in the primitive HSCs. From the latter approach, we identified TIMP-3, SFRPs, and Slit2 as candidate proteins, potentially regulating HSC homeostasis in the bone marrow niche. Further analysis will lead to a better understanding of stromal cell-mediated regulation of HSC self-renewal, proliferation, and differentiation.

3. The role of CCAAT/enhancer-binding protein ε and α in normal hematopoiesis and leukemogenesis

Yumi Fukuchi[1], Naohide Watanabe[3], Makoto Handa[3], Yasuo Ikeda[4], James N Ihle[5], Scott Kogan[6], Grant McArthur[7], Toshio Kitamura[1] and Hideaki Nakajima: [3]Deparment of Transfusion and Cellular Therapy, Keio University School of Medicine [4]Division of Hematology, Department of Internal Medicine, Keio University School of Medicine [5]Department of Biochemistry, St Jude Children's Research Hospital [6]Comprehensive Cancer Center, University of California, San Francisco [7]Division of Hematology/Medical Oncology, Peter MacCallum Cancer Institute

Granulocyte colony-stimulating factor (G-CSF) is a major cytokine that regulates proliferation and differentiation of myeloid cells, although the underlying mechanisms by which G-CSF controls myeloid differentiation are largely unknown. Differentiation of hematopoietic cells is regulated by lineage-specific transcription factors, and gene-targeting studies previously revealed the critical roles of CCAAT/enhancer-binding protein C/EBPα and C/EBPε, respectively, in the early and mid-late stages of granulocyte differentiation. The expression of C/EBPε in 32Dcl3 cells and FDCP1 cells expressing mutant G-CSF receptors was examined and it was found that G-CSF up-regulates C/EBPε. The signal for this expression required the region containing the first tyrosine residue of G-CSF receptor. Dominant-negative STAT3 blocked G-CSF-induced granulocytic differentiation in 32D cells but did not block induction of C/EBPε, indicating that these proteins work in different pathways. It was also found that overexpression of C/EBPε greatly facilitated granulocytic differentiation by G-CSF and, surprisingly, that expression of C/EBPε alone was sufficient to make cells differentiate into morphologically and functionally mature granulocytes. Overexpression of c-myc inhibits differentiation of hematopoietic cells, but the molecular mechanisms of this inhibition are not fully understood. In 32Dcl3 cells overexpressing c-myc that do not differentiate by means of G-CSF, induction of C/EBPε is completely abrogated. Ectopic expression of C/EBPε in these cells induced features of differentiation, including changes in nuclear morphologic characteristics and the appearance of granules. The data show that C/EBPε constitutes a rate-limiting step in G-CSF-regulated granulocyte differentiation and that c-myc antagonizes G-CSF-induced myeloid differentiation, at least partly by suppressing induction of C/EBPε.

Acute promyelocytic leukemia is characterized by the balanced taranslocation t(15;17), which generates PML-RARα fusion protein. This fusion protein is thought to affect key differentiation pathway of normal myeloid development, one of which is C/EBPε. We employed PML-RARα transgenic mouse model to show that restoration of C/EBPε expression can revert leukemic phenotype of these mice. These observations reveal that C/EBPε is a critical target of PML-RARα and suggest that targeted modulation of C/EBP activities could provide a new approach to therapy of AML

4. Role of C/EBPα in lineage specification, transdifferentiation and stem cell function in hematopoiesis.

Yumi Fukuchi[1], Fumi Shibata[1], Miyuki Ito, Mamoru Ito[8], Toshio Kitamura[1], Hideaki Nakajima: [8]Central Institute for Experimental Animals

CCAAT/enhancer binding protein α (C/EBPα) is a member of the bZIP family of transcription factors that plays a critical role for early myeloid differentiation. C/EBPα knockout mice showed a complete differentiation block at myeloblast stage in hematopoietic system, and mature neutrophils and eosinophils are absent in the peripheral blood. Lineage specification in developmental tree of hematopoiesis is generally

determined by lineage specific transcription factors such as C/EBPα and GATA-1 that allow commitment to the specific lineage with simultaneous extinction of their capacity to differentiate into the other ones. However, recent evidences revealed by the ectopic expression of above transcription factors unveiled the unexpected developmental plasticity of various progenitors such as MEP (erythroid/megakaryocyte progenitor) and CLP (common lymphoid progenitor). GATA-1 is reported to convert CLP and CMP (common myeloid progenitor) into erythroid/megakaryocyte lineage, however, the effect of C/EBPα on MEP and CLP is still unclear. In order to investigate the role of C/EBPα in the various aspect of hematopoietic differentiation, especially its effect on the lineage specification at different stages of differentiation in vivo, we generated transgenic mice expressing inducible form of C/EBPα (C/EBPα-ER) under H-2K promoter (C/EBPα-ER Tg). In these mice, C/EBPα activity can be induced conditionally by 4-hydroxy tamoxifen (4-HT) in all hematopoietic cells. As expected, C/EBPα-ER was expressed in almost all hematopoietic tissues including bone marrow, spleen and thymus in these mice. Gel shift analysis revealed that C/EBPα-ER was activated by 4-HT, and showed specific binding to C/EBP-specific oligonucleotide in these tissues. Next we tested differentiation plasticity of erythroid and lymphoid progenitors by ectopically inducing C/EBPα-ER activity in these cells. We sorted MEP and CLP by FACS from C/EBPα-ER Tg and examined their clonogenic activities in the presence or absence of 4-HT. In the absence of 4-HT, MEP and CLP exclusively formed erythroid/megakaryocyte and lymphoid colonies, respectively, as previously reported. Surprisingly however, these cells dramatically changed their fate of differentiation and formed significant numbers of granulocyte/macrophage (GM) colonies in the presence of 4-HT, indicating that ectopic activation of C/EBPα-ER activity skewed their differentiation pathways to myeloid lineage. Cytospin preparation of the colonies and RT-PCR analysis revealed that these were accompanied by the morphological differentiation to granulocytes/macrophages, and upregulation of myeloid-specific genes at mRNA level. These results indicate that MEP and CLP are not fully committed to either erythroid/megakaryocyte or lymphoid lineage, and possess differentiation plasticity that can be redirected to myeloid lineage.

5. Isolation and characterization of placental mesenchymal stem cells (MSCs)

Yumi Fukuchi[1], Fumi Shibata[1], Miyuki Ito, Toshio Kitamura[1], Hideaki Nakajima

MSC is a cell that has a capacity to differentiate to muscle cells, bone, cartilage, adipocytes or cardiomyocytes. MSCs are widely distributed in a variety of tissues in the adult human body as well as in the fetal environment. Since the methods for isolation, culture and differentiation induction of MSC are still not fully established for clinical application, and the more feasible, easily-obtainable source for MSC are wanted, we tried to identify cells with MSC-like potency in human placenta. We isolated adherent cells from trypsin-digested term placentas and established two clones by limiting dilution. We examined these cells for morphology, surface markers, gene expression patterns, and differentiation potential and found that they expressed several stem cell markers, hematopoietic/endothelial cell-related genes, and organ-specific genes, as determined by RT-PCR and FACS. They also showed osteogenic and adipogenic differentiation potentials under appropriate conditions. We suggest that placenta-derived cells have multilineage differentiation potential similar to MSCs in terms of morphology, cell-surface antigen expression, and gene expression patterns. These new methods of MSC isolation from human placenta will pave a way to their application in the clinical settings.

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Leading Project of the Regenerative Medicine

Laboratory of Developmental Stem Cell Biology

文部科学省　再生医療の実現化プロジェクト
幹細胞探索研究領域

Project Associate Professor　Hideo Ema M.D., Ph.D.　特任准教授　医学博士　依　馬　秀　夫

The mission of this project is to explore the basic principles in stem cell biology that can be translated into stem-cell therapy. Hematopoietic stem cells (HSCs) are capable of both self-renewal and multilineage differentiation. HSCs can clonally produce all blood cells throughout the lifetime of an organism. We have been determined to work on HSCs because these stem cells have already been used in transplantation, and are expectedly applied to a variety of clinical settings. Mouse HSCs have been an excellent stem cell model because of the existence of established methods for their functional identification. We have attempted to clarify regulatory mechanisms for mouse HSC self-renewal and differentiation.

1. Asymmetric division in HSCs

Jun Ooehara[1], Toshinobu Nishimura[1], Hiromitsu Nakauchi[1], and Hideo Ema: [1]Laboratory of Stem Cell Therapy, Center for Experimental Medicine, IMUT.

Every multicellular organism consists of a variety of different cell types. To generate this diversity, one cell forms two daughter cells that are functionally different. This is the basis for asymmetric cell division. Stem cells presumably use this manner of cell division to maintain themselves. Although to know whether this is the case for all sorts of stem cells is not easy at all, we should be able to make representative use of certain stem cells to study molecular mechanisms for asymmetric division involving both self-renewal and differentiation.

We provided evidence for asymmetric division in mouse HSCs. When single HSCs divided once in culture with cytokines, only one of the two daughter cells was shown to be detected as HSCs. In the adaptor protein Lnk-deficient mice, the number of HSCs is markedly increased due to their enhanced self-renewal capacity. We have shown that symmetrical self-renewal takes place in $Lnk^{-/-}$ HSCs, but not in wild-type HSCs in the presence of stem cell factor (SCF) and thrombopoietin (TPO). Later on we showed that Lnk negatively regulates self-renewal via TPO signal transduction pathways in HSCs.

To extend these observations and address molecular mechanisms underlying asymmetric self-renewal, we have hypothesized the basic properties of HSCs and multipotent progenitors as follows: (1) Adult HSCs undergo asymmetric division in the presence of SCF＋TPO. This system is not always reflecting in vivo phenomenon but still useful for studying molecular mechanisms of asymmetric self-renewal. (2) Multipotent progenitors undergo symmetric self-renewal in the presence of SCF＋TPO. These might sound somewhat strange. But, the pool of progenitors but not of stem cells should be remarkably expanded to sufficiently supply large numbers of non-dividing mature blood cells. In contrast, stem cells but not progenitors need to maintain their line. (3) The lifespan in terms of the number of cell cycles they can go through should

discriminate stem cells from progenitors.

We have developed a sensitive competitive repopulation assay which permits us to detect both HSCs and progenitors with limited lifespan. We performed so called paired daughter cell experiments with this new assay. We have collected data showing that one HSC can give rise two daughter cells of which one is an HSC and the other is a committed progenitor. It should be emphasized that HSCs can commit to a single lineage after only one division by losing other differentiation potentials including lymphoid ones. These data apparently contradict the currently accepted view of HSC differentiation pathways; Loss of either myeloid or lymphoid differentiation potential is the first step of lineage commitment. On the other hand, a number of multipotent progenitors can be rather easily generated in culture, supporting our hypothesis that they can undergo symmetric self-renew but only limited times at the certain differentiation stages.

On the other hand, we have noticed that the total HSC activity, as defined by repopulating units, significantly increases in HSCs by their bulk culture with SCF+TPO whereas single HSCs seemed never to undergo symmetric self-renewal under the same conditions. This evokes a possibility that the cell-cell interaction between HSCs and progenitors might support symmetric self-renewal in HSCs. This interaction can be mediated by adhesion molecules or secreted soluble proteins. Soluble proteins are more likely because direct cell-cell contact was not necessary for the increase in HSC activity.

In order to define HSCs and multipotent progenitors by molecular terms, we have been performing microarray analyses for expressed genes in HSCs and progenitors from wild type and Lnk-/- mice. Hopefully we are able to select candidate genes playing a role in cell division manners intrinsically and extrinsically or being associated with the number of cell division that cells are allowed to have.

2. Presence of alternative common myeloid progenitors

Yohei Morita[1], Jun Ooehara[2], Hiromitsu Nakauchi[2], and Hideo Ema: [1]ReproCell, [2]Laboratory of Stem Cell Therapy, Center for Experimental Medicine, IMUT.

It is very important to have the absolutely pure population of HSCs for progress in stem cell biology. To address this issue we have attempted to further fractionate $CD34^-$c-$Kit^+Sca\text{-}1^+$lineage $marker^-$ ($CD34^-$KSL) cells in the adult mouse bone marrow. We tested >100 antibodies that recognize different cell surface antigens. Most antibodies stained either all or no cells in this population, suggesting that this population is almost phenotypically homogenous, and members in the population are closely related. Nonetheless, we found that both CD150 and CD 41 expression further divide $CD34^-$KSL cells into four subpopulations. We have extensively characterized these subpopulations. $CD150^+$ cells were enriched in HSCs, and $CD150^-$ cells were enriched in HSCs with significantly less self-renewal capacity, supporting previous observations. Interestingly, $CD150^+CD41^-$ but not $CD150^+CD41^+$ cells appeared enriched in HSCs. $CD150^+CD41^+$ cells were still able to form in vitro myeloid colonies many of which consisted of all neutrophils, macrophages, erythroblasts, and megakaryocytes (nmEM colonies). The originally reported common myeloid progenitors (CMPs) with the Sca-1^-c-$Kit^+Lin^-CD34^+FcgR^{lo}$ phenotype cannot make nmEM colonies (Our unpublished data). In this regard, $CD150^+CD41^+CD34^-$ KSL cells may represent true CMPs closely related to HSCs. Otherwise, some of myeloid as well as lymphoid differentiation potentials can be directly lost, perhaps, at a random fashion, from HSCs as a first event of lineage commitment.

3. Wnt3a is not a self-renewal factor for HSCs

Satoshi Yamazaki[1], Akira Kikuchi[2], Hiromitsu Nakauchi[3], and Hideo Ema: [1]ReproCell, [2]Department of Biochemistry, Graduate School of Biomedical Sciences, Hiroshima University, [3]Laboratory of Stem Cell Therapy, Center for Experimental Medicine, IMUS.

Wnt3a reportedly acts as a self-renewal factor on mouse HSCs. We have made efforts to confirm this finding because it is extremely important for developing new stem cell therapy using HSCs. It was reported that the purity of Wnt protein is crucial to induce self-renewal of HSCs. Moreover, lipid modifications have to be maintained during its purification processes. We had spent years to obtain highly purified and biologically active mouse Wnt3a protein without success.

Recently Dr. Kikuchi, Hiroshima University et al. have successfully purified mouse Wnt3a and Wnt5a in which biological activity is retained. We immediately started our collaboration. We detected Wnt3a activity using mouse HSCs in the following two ways. After stimulation with Wnt3a, β-catenin was localized into the nucleus in HSCs. After stimulation with Wnt3a, Laz activity was detected in HSCs from TOP-Gal

transgenic mice. However, unlike previous studies, this study showed that Wnt3a does not either induce or enhance self-renewal in HSCs at all. To reproduce these results, we have been performing transplantation experiments once again.

We have made interesting observations during this study. Translocalization of β-catenin occurred in HSCs after stimulation with cytokines such as TPO and SCF. The significance of β-catenin signal in HSCs is under investigation. On the other hand, the expression of Wnt3a cannot be detected in any adult tissues. The development of HSCs in the fetal liver is severely impaired in Wnt3a-deficient mice. TOP-EGFP transgenic mice should be useful for studying when and where Wnt3a plays a role in HSC development. We hope that this study will clarify a role of Wnt3a in the regulation of HSCs.

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Leading Project of the Regenerative Medicine

Laboratory of Stem Cell Regulation

文部科学省　再生医療の実現化プロジェクト
幹細胞制御領域

Project Associate Professor　Koichi Hattori, M.D., Ph.D.
Project Assistant Professor　Beate Heissig, M.D.

特任准教授　医学博士　服　部　浩　一
特任助教　医学博士　ハイズィーッヒ･ベアーテ

The major goal of our laboratory is to understand how stem cells are regulated and how we can apply this knowledge for stem-cell based regenerative medicine. Stem cells can differentiate into tissue-specific cells to repair damaged tissue, a process controlled in part by the microenvironment. Proteases, as part of the microenvironment act as processing enzymes that perform highly selective and limited cleavage of specific substrates including growth factors and their receptors, cell adhesion molecules, cytokines, chemokines, apoptotic ligands and angiogenic factors. Over the last year we studied the role of the plasminogen fibrinolytic pathway for hematopoietic regeneration (1) and the role of proteases in neurotoxicity of amyotrophic lateral sclerosis (2).

1) The Plasminogen Fibrinolytic Pathway Is Required for Hematopoietic Regeneration

Beate Heissig, Leif R. Lund[1], Haruyo Akiyama, Makiko Ohki, Yohei Morita, John Rømer[1], Hiromitsu Nakauchi, Ko Okumura[2], Hideoki Ogawa[2], Zena Werb[3], Keld Danø[1], and Koichi Hattori: [1]Finsen Laboratory, Rigshospitalet, Strandboulevarden 49, 2100 Copenhagen, Denmark [2]Atopy (Allergy) Research Center, Juntendo University School of Medicine, 2-1-1 Hongo, Bunkyo-ku, Tokyo, 113-8421, Japan [3]Department of Anatomy, University of California, San Francisco, 513 Parnassus Avenue, San Francisco, CA 94143-0452, USA.

Hematopoietic stem cells within the bone marrow exist in a quiescent state. They can differentiate and proliferate in response to hematopoietic stress (e.g., myelosuppression), thereby ensuring a well-regulated supply of mature and immature hematopoietic cells within the circulation. However, little is known about how this stress response is coordinated. Here, we show that plasminogen (Plg), a classical fibrinolytic factor, is a key player in controlling this stress response. Deletion of Plg in mice prevented hematopoietic stem cells from entering the cell cycle and undergoing multilineage differentiation after myelosuppression, leading to the death of the mice. Activation of Plg by administration of tissue-type plasminogen activator promoted matrix metalloproteinase-mediated release of Kit ligand from stromal cells, thereby promoting hematopoietic progenitor cell proliferation and differentiation. Thus, activation of the fibrinolytic cascade is a critical step in regulating the hematopoietic stress response.

2) Matrix metalloproteinase-9 regulates TNF-alpha and FasL expression in neuronal, glial cells and its absence extends life in a transgenic mouse model of amyotrophic lateral sclerosis.

Mahmoud Kiaei[1], Khatuna Kipiani[1], Noel Y.

Calingasan[1], Elizabeth Wille[1], Junyu Chen[1], Beate Heissig, Shahin Rafii[2], Stefan Lorenzl[1] and M. Flint Beal[1]: [1]Department of Neurology and Neuroscience, Weill Medical College of Cornell University, 525 East 68th Street, New York, NY 10021, USA [2]Department of Medicine, Weill Medical College of Cornell University, New York-Presbyterian Hospital, New York, NY 10021, USA

Whether increased levels of matrix metalloproteinases (MMPs) correspond to a role in the pathogenesis of amyotrophic lateral sclerosis (ALS) needs to be determined and it is actively being pursued. Here we present evidence suggesting that MMP-9 contributes to the motor neuron cell death in ALS. We examined the role of MMP-9 in a mouse model of familial ALS and found that lack of MMP-9 increased survival (31%) in G93A SOD1 mice. Also, MMP-9 deficiency in G93A mice significantly attenuated neuronal loss, and reduced neuronal TNF-alpha and FasL immunoreactivities in the lumbar spinal cord. These findings suggest that MMP-9 is an important player in the pathogenesis of ALS. Our data suggest that the mechanism for MMP-9 neurotoxicity in ALS may be by upregulating neuronal TNF-alpha and FasL expression and activation. This study provides new mechanism and suggests that MMP inhibitors may offer a new therapeutic strategy for ALS.

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Center for Asian Infectious Diseases

IMSUT Research Center for Infectious Diseases in China

中国における感染症研究拠点

Professor	Aikichi Iwamoto, M.D., D.M.Sc.	教授(兼)	医学博士	岩 本 愛 吉
Professor	Yoshihiro Kawaoka, D.V.M., Ph.D.	教授(兼)	獣医学博士	河 岡 義 裕
Project Professor	Kunito Yoshiike, D.Sc.	特任教授	理学博士	吉 池 邦 人
Project Professor	Mitsue Hayashi, Ph.D.	特任教授	人類学博士	林 光 江
Project Professor	Yoshihiro Kitamura, M.D., Ph.D.	特任教授	医学博士	北 村 義 浩
Project Associate Professor	Zene Matsuda, M.D., Ph.D, D.Sc.	特任准教授	医学博士	松 田 善 衛
Project Assistant Professor	Fumihiro Yagyu, Ph.D.	特任助教	医学博士	柳 生 文 宏

The Institute of Medical Science, University of Tokyo (IMSUT) has established Japan-China joint laboratories for research on emerging and re-emerging infectious diseases in Asia, in collaboration with the Chinese Academy of Sciences and Chinese Academy of Agricultural Sciences. In the laboratories, Japanese and Chinese scientists conduct research on the viral pathogenicity, the genetic variation of viruses in the field, structure of viral and host proteins, and so on.

BACKGROUND

Historically, China is a very important neighbor of Japan. Official diplomatic delegations were first sent from Japan during the Sui dynasty some 1400 years ago. Since late 20th century, geopolitical and economical interdependence between Japan and China has developed substantially and will deepen further in the future. China is an enormous country often symbolically referred to as the dragon. While China is developing and transforming rapidly in the coastal regions, its rural areas have been left far behind. With regard to infectious diseases, China is beset with problems ranging widely from those of a developing country to those of dense urban environments. No one can discuss emerging and re-emerging infectious diseases (ERID) without mentioning China. Severe acute respiratory syndrome (SARS) emerged in Guangdong and shocked the world in 2003. With Lake Qinghai as a reference point, avian influenza expanded westward in the Eurasian continent in 2005 and reached Africa in February 2006. The carrier rate of hepatitis viruses is very high and HIV infection is rapidly increasing.

Given these situations, academic collaboration on research in infectious diseases would be beneficial to both countries, facilitate mutual understanding, and help strengthen the stable long-term relationship between the two peoples. Establishing joint research laboratories in China is particularly important because this would allow Japanese scientists access to possible emerging pathogens and to have an opportunity to fight against possible emerging infections. Supported by a contract research fund from the Ministry of Education, Culture, Sports, Science and Technology (MEXT) (*Japan-China Collaboration on Emerg-*

ing and Re-emerging infectious Diseases; MEXT Project Director: Aikichi Iwamoto), IMSUT has established two joint laboratories in Beijing in collaboration with the Institute of Biophysics and Institute of Microbiology, Chinese Academy of Sciences (IBPCAS and IMCAS, respectively); a collaborative research program with Harbin Veterinary Research Institute (HVRI), the Chinese Academy of Agricultural Science; and IMSUT's project office in Beijing.

LABORATORIES AND PROJECT OFFICE

a. Laboratory of Structural Virology and Immunology (LSVI)

In May 2006, LSVI (PI: Z. Matsuda) was established on the third floor of IBP Building 1. IBP is one of the leading institutes for protein sciences and structural biology in China and is now under new Director Tao Xu. The laboratory's physical environment has been greatly improved during 2007, reaching the final stage of the initial plan. Furthermore, the laboratory personnel increased gradually. In 2007, Naoyuki Kondo, a Japanese crystallographer, joined LSVI from Osaka University. LSVI has successfully recruited three Chinese researchers this year and the total number of laboratory staff became nine. To take advantage of being located in IBP, they are trying to build a bridge between protein science and virology. They have set the primate lentiviruses (HIV and SIV) as research targets, and they are analyzing functions of lentiviral as well as relevant host proteins with a strong emphasis on structural analyses using X-ray crystallography and other techniques. They are also employing a protein engineering technology to generate versatile reporter proteins to monitor fundamental biological events mediated by viral proteins, such as membrane fusion.

b. Laboratory of Molecular Immunology and Molecular Microbiology (LMIMM)

LMIMM, founded in May 2006, was set to work by Y. Kitamura, PI, and his associates in October 2006 in IMCAS. In January 2007, LMIMM moved to an IM's new building in the CAS Olympic Science Park. They have re-established LMIMM, with increased laboratory staff and facilities: the laboratory is now equipped with basic apparatus for molecular studies and rooms for virus experiments and for bacterial experiments, required for their research. Chie Aoki joined LMIMM in April 2007 and F. Yagyu, an epidemiologist, in July 2007. LMIMM scientists are currently focusing on hepatitis C virus (HCV) and human immunodeficiency virus (HIV), to obtain a better understanding of mechanisms of the viral replication. Using the human cell line-infectious HCV RNA system, they are characterizing the adapted virus population and in particular, are testing whether some host genes stimulated by interferon have anti-HCV functions. To investigate the pathogenicity of the viruses in the field, they are analyzing clinical samples from HIV-infected individuals and chronic hepatitis C patients being treated in some collaborating hospitals in Beijing.

c. Collaborative research program with HVRI

HVRI (Director, Xiangang Kong) serves as a research center that focuses on the study of avian influenza viruses in domestic fowl and wild waterfowl in China. Japanese scientists from IMSUT (headed by Y. Kawaoka), together with Chinese scientists at HVRI (headed by Hualan Chen), analyzed the pathogenesis of an H5N1 influenza virus, isolated from a human in China, in rhesus macaques at a BSL3 laboratory at HVRI. They showed that this H5N1 virus replicated in the lower respiratory tract of rhesus macaques, causing severe lung damage. Since the target of H5N1 avian influenza virus in humans is thought to be the lower respiratory tract, rhesus macaques may provide a useful model to study the disease pathogenesis of H5N 1 virus in humans. They are also carrying out surveillance programs and studying genetic changes in avian influenza viruses isolated in Asian countries.

d. IMSUT Project Office

The office (M. Hayashi and K. Yoshiike) is supporting the activities of the two joint laboratories in Beijing and one program in Harbin. It serves as Secretariat for Steering Committee Meeting and files MOU and Minutes. It helps scientists visiting the joint laboratories and program for collaborative research.

IMPLEMENTATION OF COLLABORATION

The collaboration is implemented, being based on MOU and Minutes of Meeting between IMSUT and the Chinese institutes. For the joint laboratories the implementation is being controlled by the steering committee consisting of Dean of IMSUT (Motoharu Seiki), MEXT Project Director and Directors of IBPCAS (Tao Xu) and IMCAS (George Fu Gao). The collaborative program in Harbin is being implemented by the steering committee consisting of Dean of IMSUT, Director of HVRI, H. Chen and Y. Kawaoka.

Publications

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