

## 2.4 DIVISION OF APPLIED BIOSCIENCES

The Division of Applied Biosciences was created in 1996 to consolidate the three divisions of Agricultural Biology (founded in 1953), Fisheries Science (1953), Animal Science (1976) and two research laboratories from Division of Tropical Agriculture (1981).

The division is conducting the research and education on the subjects aiming at the efficient utilization, conservation and creation of species for a variety of living organisms including microorganisms, plants, animals and fishes living in land and sea from the molecular to population levels. The division consists of 17 laboratories in which 109 master students including 3 students from abroad and 64 Ph D. students including 10 students from abroad are enrolled.

### Chair of Science of Plant Resources

#### 2.4.1 Laboratory of Plant Genetics

*Staff*      *Professor*                      : Endo, Takashi, D. Agric. Sci  
                 *Associate Professor* : Miyashita, Naohiko, Ph. D.  
                 *Assistant Professor* : Nasuda, Shuhei, Ph. D.

*Students and research fellows*

*Doctor's program* : (2)              *JSPS Postdoctoral Fellow* : (1)  
*Master's program* : (5)              *Undergraduate* : (2)

### A. Research Activities (2006.4-2007.3)

#### A-1. Main Subjects

##### a) Cytogenetic analysis of a genetic genome rearrangement system in wheat

Chromosomal structural changes frequently occur in specific lines of common wheat carrying certain alien chromosomes from wild species related to wheat. These chromosomal aberrations can be identified by the chromosome banding and *in situ* hybridization techniques. Using this genome rearrangement system, we have established deletion and translocation lines of wheat and are conducting studies on chromosome mapping and the introduction of useful genes from alien species, such as barley and rye, into bread wheat. Also, we have initiated a basic study aimed at cloning the gene responsible for the unique genome rearrangement system.

##### b) Molecular cytogenetic analysis of wheat and its relative species

We have been trying to shed light on the structure of the three wheat genomes by employing various molecular cytogenetic techniques. In order to characterize specific regions of wheat chromosomes efficiently, we have developed a high-throughput marker system derived from the AFLP (amplified fragment length polymorphism) technique. We have also been conducting molecular cloning experiments aimed at isolating genes encoding for centromeric proteins in wheat to elucidate the structure and function of the plant centromeres. A major characteristic of wheat genomes is the abundance of repetitive elements. To reveal their structure and evolutionary consequence in the genomes of wheat and its relatives, we are trying to clone and

analyse species-specific repetitive elements.

- c) Molecular population genetics study on DNA variation in the genera *Arabidopsis* and *Arabis*, and *Oryza*.

To establish a model system for plant population genetics, we have chosen two genera *Arabidopsis* and *Arabis*. So far, several genic regions have been analyzed to quantify DNA variation at a specific genic region. In addition, microsatellite and AFLP analyses were conducted to investigate the pattern of DNA polymorphism over the entire genome. Currently, we are analyzing genes involved in herbicide resistance in *A. thaliana*. To examine a more general picture of molecular variation in plant species, we are planning to compare these two genera and *Oryza* species, which are monocots. We have already analyzed DNA variation in two *Adh* locus regions in *Oryza* species, and are analyzing Blast-related and flowering genes.

## A-2. Publications and presentations

- a) Publications

### *Original papers*

- Rakshit, S., A. Rakshit, H. Matsumura, Y. Takahashi, Y. Hasegawa, A. Ito, T. Ishii, N. T. Miyashita and R. Terauchi, 2007 Large-scale DNA polymorphism study of *Oryza sativa* and *O. rufipogon* reveals the origin and divergence of Asian rice. *Theor. Appl. Genet* 114: 731-743
- Kawabe, A., Nasuda, S., Charlesworth, D.: Duplication of centromeric histone H3 (HTR12) gene in *Arabidopsis halleri* and *lyrata*, plant species with multiple centromeric satellite sequences. *Genetics* 174:2021-2032, 2006
- Kawabe, A. and Nasuda, S.: Chromosome-specific satellite sequences in *Turritis glabra*. *Genes Genet. Syst.* 81:287-290, 2006
- Kawabe, A. and Nasuda, S.: Polymorphic chromosomal specificity of centromere satellite families in *Arabidopsis halleri* ssp. *gemmifera*. *Genetica* 126:335-342, 2006
- Masoudi-Nejad, A., Tonomura, K., Kawashima, S., Itoh, M., Kanehisa, M., Endo, T., and Goto, S.: EGAssembler: online bioinformatics service for large-scale processing, clustering and assembling ESTs and genomic DNA fragments. *Nucleic Acids Research*. doi:10.1093/nar/gkl066, 2006
- Suchánková, P., Kubaláková, M., Kovářová, P., Bartoš, J., Číhalíková, J., Molnár-Láng, M., Endo, T.R., and Doležel, J.: Dissection of the nuclear genome of barley by chromosome flow sorting. *Theor. Appl. Genet.* (on line) DOI 10.1007/s0012-006-0329-8, 2006

- b) Conference and seminar papers presented

The 78<sup>th</sup> Annual meeting of the Genetic Society of Japan: 3 presentations

The 109<sup>th</sup> annual meeting of the Japanese Society of Breeding: 1 presentations

The 110<sup>th</sup> annual meeting of the Japanese Society of Breeding: 1 presentation

## A-3. Off-campus activities

### *Membership in academic societies*

Endo, T.: Genetic Society of Japan (Editor-in-Chief)

Endo, T.: Genetic Resources Committee and Resource Center, National Institute of Genetics (Member)

### ***Research grants***

Grant-in-Aid for Scientific Research from the Ministry of Education, Science, Sports and Culture  
(C) (Nasuda)

National Bioresource Project (the Ministry of Education, Science, Sports and Culture) (Endo and Nasuda)

## **A-4. International cooperations and overseas activities**

### ***International joint research, overseas research surveys***

Nasuda, S., Endo, T. R.: Joint Research Project, Japan-Germany Research Cooperative Program, JSPS. "Identification of the nucleotide sequences necessary for functional centromeres in barley"

### ***Scholars from abroad***

Collaborative researcher: from Australia (1)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### **a) Courses given**

Undergraduate level: Basic Bioresource Science I (Endo), Outline of Bio-production Science I (Endo), Genetics I (Endo), Genetics II (Miyashita), laboratory of Bioresource Science I, II (Endo and Miyashita), Seminar in Plant Resource Science (Endo and Miyashita)

Graduate level: Genetics (Advance Course) I (Endo), Genetics (Advance Course) II (Miyashita), Seminar in Plant Genetics (Endo and Miyashita), Research in Plant Genetics (Endo and Miyashita)

### **B-2. Off-campus teaching, etc**

#### ***Part-time lecturer***

Nasuda, S.: Faculty of Education, Kyoto University of Education (Genetics)

Nasuda, S.: Doshisha Women's College of Liberal Arts (Life Sciences)

## 2.4.2 Laboratory of Crop Evolution

*Staff*      *Professor*                      : Ohnishi, Ohmi, Ph.D.  
                 *Associate professor* : Kawahara, Taihachi, Dr. Agr. Sci.  
                 *Instructor*                        : Yasui, Yasuo, Dr. Agr. Sci.  
                 *Research associate* : Imai, Takehiro, Dr. Bio. Engi.

*Students and research fellows:*

*Doctor's program:* (3)

*Master's program:* (2)

### A. Research Activities (Jan. 2006-Dec. 2006)

#### A-1. Main subjects

##### a) Studies on the origin and evolution of cultivated plants

**Emmer wheat:** The domestication of emmer wheat was one of the key events during the emergence of agriculture in southwestern Asia, and was a prerequisite for the evolution of durum and common wheat. Genotypes based on restriction fragment length polymorphism were analyzed to describe the structure of populations of wild and domesticated emmer and to generate a picture of emmer domestication and its subsequent diffusion across Asia, Europe and Africa. Wild emmer consists of two populations, southern and northern, each further subdivided. Domesticated emmer mirrors the geographic subdivision of wild emmer into the northern and southern populations and also shows an additional structure in both regions. Gene flow between wild and domesticated emmer occurred across the entire area of wild emmer distribution. Emmer was likely domesticated in the Diyarbakir region in southeastern Turkey, followed by subsequent hybridization and introgression from wild to domesticated emmer in southern Levant. Durum wheat is closely related to domesticated emmer in the eastern Mediterranean and likely originated there.

**Buckwheat:** In this year, search for the wild ancestral (*Fagopyrum esculentum* ssp. *ancestrale*) populations and populations of closely related self-pollinated wild species, *F. homotropicum* was conducted in the Sichuan-Yunnan border area of China. The Tongyi river valley was confirmed to be the area where many large natural populations of the wild ancestor are growing. In addition, the Niru river valley of Shangurila district of Yunnan province was revealed to be the other area where the wild ancestor is growing as many large populations. Allozyme analysis of these population revealed that the natural populations of the wild ancestor from these valleys were not closely related to cultivated populations of common buckwheat. The original birthplace of cultivated common buckwheat is the Langcan river (upper stream of the Mekong river) valley of Yunnan-tibet border area, not to be the Tongyi river valley. In the Tongyi river valley and the Niru river valley, diploid form of *F. cymosum* is growing, suggesting that the Tongyi river valley is the birthplace of the wild ancestor itself. We need further experimental analyses on this account.

The issue of gene flow between the wild ancestor and cultivated common buckwheat was tackled by using microsatellite markers which were recently developed by Konishi et al. (2006). The gene flow between distinct cultivated populations was detected, but the gene flow between the wild ancestor and cultivated buckwheat and between distinct natural populations of the wild

ancestor was negligible as the source of genetic variation.

Radish: Phylogenetic relationships among cultivars and natural populations of wild radish species have been analyzed using genetic variations of chloroplast DNA. The same issue was tackled by analyzing PCR-RFLP variations of nuclear genes. The phylogenetic relationships among them were the same as those revealed by cpDNA analyses. This implies that our previous results on phylogenetic relationship based on cpDNA and the result newly obtained were strictly true. Our further analyses of genetic variations from the viewpoint of the origin of cultivated radish will be pursued based on these results on phylogenetic relationships among cultivars and natural populations.

Natural populations of wild radish, *Raphanus raphanistrum*, were analyzed for their maintenance of genetic variation using allozyme variations. Furthermore, phylogenetic relationships among them were also analyzed by using allozymes and chloroplast DNA variations.

b) Studies on the evolution of wheat and its wild relatives

*Ae. caudata*: Variation of *Aegilops caudata*, the wild relative of wheat with C genome was surveyed by using cpDNA SSR regions. We analyzed 20 SSR regions in 96 accessions of *Ae. caudata* and found out two distinct haplogroup in this species. One group was found in the whole distribution area of *Ae. caudata* but the other was found only in the western part of its geographical range, Western Turkey and Greece. Since hybrid sterility has been reported in this species, detailed phylogenetic analysis is needed to understand the evolution of the two groups.

c) Studies on the self-incompatibility gene of common buckwheat

In common buckwheat, the self-incompatibility gene is closely linked with the genes controlling several morphologies related with heterostyly and they form the S supergene. So far, the linkage map and genomic library have been constructed as a first step for the cloning of the S gene by the positional cloning method. In this year, total DNA were extracted from 4800 individual and the map distance between S gene and the molecular marker obtained was estimated to 0/4800 cM. Screening of the genomic library and chromosome walking is now under progress.

d) Screening of useful genes in plant genetic resources

Screening of basic useful characteristics is indispensable for utilization of plant genetic resources. We checked spring vs. winter growing habit in 164 accessions of *Ae. umbellulata*. About one fourth of the accessions was spring growth habit, headed within 50 days under continuous light with minimum of 15 degrees. Another one fourth was winter growth habit and did not headed within 100 days. We found several accessions with very early and spring growing habit and started genetic analysis in these.

## A-2. Publication and presentation

a) Publication

**Original papers**

Dudnikov, A. J. and T. Kawahara: *Aegilops tauschii*: genetic variation in Iran. Genetic Resources and Crop Evolution. 53: 579-586. 2006

Fawcett, J. A., T. Kawahara, H. Watanabe and Y. Yasui: A SINE family widely distributed in the plant kingdom and its evolutionary history. Plant Molecular Biology. 61: 505-514. 2006

Kawasaki, M. and O. Ohnishi: Two distinct groups of natural populations of *Fagopyrum urophyllum* (Bur. et. Franch.) Gross revealed by the nucleotide sequence of a noncoding

- region in chloroplast DNA. *Genes & Genetic Systems* 81: 323-332. 2006
- Konishi, T., H. Iwata, K. Yashiro, Y. Tsumura, R. Ohsawa, Y. Yasui and O. Ohnishi: Development and characterization of microsatellite markers for common buckwheat. *Breeding Science* 56: 277-285. 2006
- Konishi, T. and O. Ohnishi: A linkage map for common buckwheat based on microsatellite and AFLP markers. *Fagopyrum* 23: 1-6. 2006
- Onishi, A. T. Hongo, T. Sasakuma, T. Kawahara, K. Kato and H. Miura: Variation and segregation for rachis fragility in spelt wheat, *Triticum spelta* L. *Genetic Resources and Crop Evolution*. 53: 985-992. 2006
- Yamane, K., K. Yano, and T. Kawahara: Pattern and Rate of Indel Evolution Inferred from Whole Chloroplast Intergenic Regions in Sugarcane, Maize and Rice. *DNA Research* 13: 197-204. 2006
- b) Conference and seminar papers presented
- 109th, 110th annual meeting of the Japanese Society of Breeding: 4 papers
- Annual meeting of the Japanese Society of Genetics in 2006: 2 papers

### **A-3 Offcampus activities**

#### ***Membership of academic societies (roles)***

- Ohmi Ohnishi and Taihachi Kawahara: Kihara Memorial Yokohama foundation for the Advancement of Life Sciences (Visiting researchers)
- Ohmi Ohnishi: Japanese Society of Breeding (A member of Editorial Board)

#### ***Research Grant***

- National Bioresource Project KOMUGI (the Ministry of Education, Science, Sports and Culture) (Kawahara, Yasui)
- Grant-in-Aid for Scientific Research from the Japanese Society for Promotion of Science (A): Analysis of genetic diversity in plants with wide distribution in Eurasia by using wild relatives of wheat (Kawahara, Yasui)
- Grant-in-Aid for Scientific Research from the Japanese Society for Promotion of Science (B): Molecular cloning and evolutionary analysis of the heterostylous self-incompatibility gene of buckwheat (Ohnishi, Yasui)

### **A-4. International cooperation and oversea activities**

#### ***Membership in international academic societies***

- Ohnishi, O.: Editor-in-Chief of FAGOPYRUM (International journal of IBRA), A member of the editorial board of Genetic Resources and Crop Evolution (the Netherlands)

#### ***International joint Researches, oversea research surveys***

- Kawahara, T.: Studies on the origin of Emmer wheat (USA)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-Campus teaching**

#### **a) Courses given**

- Undergraduate level: Plant Genetic Resources (Ohnishi, Yasui)
- Graduate level: Origin of Cultivated Plants (Ohnishi), Ethnobotany (Kawahara)
- Seminar in Origin of Cultivated Plants (Ohnishi, Kawahara,)

## **B-2. Off-campus teaching, etc.**

### ***Part-time lecturer***

Ohnishi, O.: Grad. School of Agr. Kyoto Prefectural Univ. (Plant Genetic Resources), Fac. Agr.  
Kobe Univ. (Population genetics)

Kawahara, T.: Fac. Agr. Kyoto Prefectural Univ. (Genetics)

## **B-3. Oversea teaching**

### ***Students and research fellows from abroad***

Graduate students (2) from China and Nepal

# **Chair of Science of Plant Protection**

## **2.4.3 Laboratory of Plant Pathology**

*Staff*      *Professor*                      : Okuno, Tetsuro, Dr. Agric. Sci.  
                 *Associate Professor* : Mise, Kazuyuki, Dr. Agric. Sci.  
                 *Lecturer*                         : Takano, Yoshitaka, Dr. Agric. Sci.  
                 *Assistant Professor* : Kaido, Masanori, Dr. Agric. Sci.

### ***Students and research fellows***

*Doctor's Program* : (4)                      *Undergraduate:* (4)  
*Master's Program* : (12)

## **A. Research Activities (2006.4-2007.3)**

### **A-1. Main subjects**

#### **a) Studies on plant RNA virus infection**

Plant viruses cause serious diseases in many important plant species, but few effective antiviral strategies have been developed. This is likely due to our inadequate understanding of the basic biology of plant viruses. We are studying the mechanisms of infection and replication of plant viruses and their interaction with host plants. Viruses used for the studies are dianthoviruses and bromoviruses. Plants used for the studies are *Arabidopsis*, barley, cowpea, tobacco and others. We have been analyzing the molecular structures and the functions of viral RNAs and proteins in the infection process of viruses in both plant and protoplast systems. Researches include investigations on the early events of infection to the late maturation stage such as protein synthesis, RNA replication, the cell-to-cell movement of virus, and packaging of viral RNAs into virions and analyses of molecular mechanisms of symptom expression caused by plant virus infection.

b) Identification of host factors involved in plant virus infection

Viruses utilize host proteins to establish infection in plants. Plant virus genomes and their encoded proteins are proposed to have physical interactions with putative host-derived factors. Surveys for plant genes involved in RNA virus infections have been progressing by using proteomics approach including affinity chromatography purification and mass spectrometry and genetic approach using *Arabidopsis* mutants.

c) Studies on infection mechanism of phytopathogenic fungi

Plant pathogenic fungi cause serious diseases on a wide range of crops and ornamental plants. For development of novel strategies for protecting plants from fungal infection, it is necessary to understand both of fungal infection and plant resistant mechanisms at molecular level. Researchers are studying infection mechanism of a plant pathogenic fungus *Colletotrichum lagenarium*, the causal agent of cucumber anthracnose. Molecular genetic analysis has identified many genes involved in pathogenicity of *C. lagenarium*. Based on information about identified pathogenicity-related genes, we are especially focusing on relations of cellular signaling pathways and peroxisomal metabolic function with fungal infection mechanism. Using *Arabidopsis thaliana* and several *Colletotrichum* species that have different host ranges, we are studying nonhost plant resistance and suppression of plant defense responses by adapted pathogens.

## A-2. Publications and presentations

a) Publications

### **Books**

Okuno, T.: Mechanism of RNA silencing suppression in dianthoviruses in “Pathogenicity of Microorganisms and Plant Defense Responses” ed., I. Uyeda. Hokkaido University Press, Sapporo, p. 203-213, 2007

Okuno, T. and H. Mizumoto: Replication of *Red clover necrotic mosaic virus* in “Recent Advances in RNA Virus Replication” ed., K. L. Hefferon. Transworld Research Network, Kerala, India, p. 247-264, 2006

Mise, K.: Host Factors Involved in the Replication and Movement of *Brome mosaic virus* in “Pathogenicity of Microorganisms and Plant Defense Responses” ed., I. Uyeda. Hokkaido University Press, Sapporo, p. 173-180, 2007

Mise, K.: Virus in “Microbiology” ed., K. Aoki. Kagaku Dojin, Kyoto, p. 39-41, 2007

Mise, K.: Pathogenic Microorganisms in “Microbiology” ed., K. Aoki. Kagaku Dojin, Kyoto, p. 44-50, 2007

### **Original papers**

Kaido, M., Y. Inoue, Y. Takeda, K. Sugiyama, A. Takeda, M. Mori, A. Tamai, T. Meshi, T. Okuno and K. Mise: Downregulation of the *NbNACa1* gene encoding a movement-protein-interacting protein reduces cell-to-cell movement of *Brome mosaic virus* in *Nicotiana benthamiana*. Mol. Plant-Microbe Interact. 20(6); 671-681, 2007

Matsumoto, K., T. Toyooka, C. Tomikawa, A. Ochi, Y. Takano, N. Takayanagi, Y. Endo and H. Hori: RNA recognition mechanism of eukaryote tRNA (m<sup>7</sup>G46) methyltransferase (Trm8-Trm82 complex). FEBS Lett. 581(8); 1599-1604, 2007

Asakura, M., T. Okuno and Y. Takano: Multiple contributions of peroxisomal metabolic function to fungal pathogenicity in *Colletotrichum lagenarium*. Appl. Environ. Microbiol. 72(9); 6345-6354, 2006



- Mizumoto, H., H.-O. Iwakawa, M. Kaido, K. Mise and T. Okuno: Cap-independent translation mechanism of *Red clover necrotic mosaic virus* RNA2 differs from that of RNA1 and is linked to RNA replication. *J. Virol.* 80(8); 3781-3791, 2006
- Mori, M., H. Kitamura, A. Kondo, K. Dohi, M. Mori, M. Kaido, K. Mise, E. Shimojyo and Y. Hashimoto: Expression of an enhancin gene from the *Trichoplusia ni* granulosis virus confers resistance to lepidopterous insect pests to rice. *Plant Biotechnol.* 23(1); 55-61, 2006
- Shimada, C., V. Lipka, R. O'Connell, T. Okuno, P. Schulze-Lefert and Y. Takano: Nonhost resistance in *Arabidopsis-Colletotrichum* interactions acts at the cell periphery and requires actin filament function. *Mol. Plant-Microbe Interact.* 19(3); 270-209, 2006
- Takano Y., N. Takayanagi, H. Hori, Y. Ikeuchi, T. Suzuki, A. Kimura and T. Okuno: A gene involved in modifying transfer RNA is required for fungal pathogenicity and stress tolerance of *Colletotrichum lagenarium*. *Mol. Microbiol.* 60(1); 81-92, 2006
- Takao, Y., K. Mise, K. Nagasaki, T. Okuno and D. Honda: Complete nucleotide sequence and genome organization of a single-stranded RNA virus (SssRNAV) infecting the marine fungoid protist *Schizochytrium* sp. *J. Gen. Virol.* 87(3); 723-733, 2006
- Watanabe, T., A. Takeda, T. Tsukiyama, K. Mise, T. Okuno, H. Sasaki, N. Minami and H. Imai: Identification and characterization of two novel classes of small RNAs in the mouse germline: retrotransposon-derived siRNAs in oocytes and germline small RNAs in testes. *Genes Dev.* 20(13); 1732-1743, 2006

### **Reviews**

- Okuno, T. and A. Takeda: Virus and RNA silencing in "Responses to Environmental and Biotic Stress in Plants" eds. K. Shimamoto *et al.*, Tanpakushitsu Kakusan Koso. 52(6); 692-697, 2007
- Okuno, T.: Infection strategies of plant viruses in "Molecular Strategies in the Interaction between Pathogens and Host Plants" eds. S. Arase and M. Ueno, Proceedings of PSJ Plant-Microbe Interactions. Vol. 42; 157-167, 2006
- Okuno, T.: A research life with RNA viruses over a long way around. *RNA Network Newsletter.* 5(1); 28-30, 2006
- Takano, Y.: Molecular mechanisms of nonhost resistance in "Responses to Environmental and Biotic Stress in Plants" eds. K. Shimamoto *et al.*, Tanpakushitsu Kakusan Koso. 52(6); 705-710, 2007

### **b) Conference and seminar papers presented**

- Annual Meeting of the Phytopathological Society of Japan: 9 presentations
- Kansai Meeting of the Phytopathological Society of Japan: 2 presentations
- RNA Meeting of the RNA Society of Japan: 1 presentation

## **A-3. Off-campus activities**

### **Membership in academic societies**

- Okuno, T.: The Phytopathological Society of Japan (Councilor), The Japanese Society for Virology (Councilor), The Molecular Biology Society of Japan, The RNA Society of Japan
- Mise, K.: The Phytopathological Society of Japan, The Japanese Society for Virology, The Molecular Biology Society of Japan
- Takano, Y.: The Phytopathological Society of Japan, The Molecular Biology Society of Japan

Kaido, M.: The Phytopathological Society of Japan

#### ***Research grants***

Grant-in-Aid for Scientific Research from the Japan Society for the Promotion of Science:  
Grant-in-Aid for Scientific Research (A) (Okuno, head), Grant-in-Aid for Scientific Research (B) (Mise, head), Grant-in-Aid for Scientific Research (B) (Kaido, member)

Grant-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science and Technology, Japan: Grant-in-Aid for Scientific Research in Priority Areas (Okuno, head), Grant-in-Aid for Scientific Research in Priority Areas (Takano, member), Grant-in-Aid for Young Scientists (B) (Kaido, head)

Others: The 21st century COE program for Microbial-Process Development Pioneering Future Production Systems from the Ministry of Education, Culture, Sports, Science and Technology, Japan (Okuno, Mise, member), Investigation of Research Development in Agricultural Sciences from the Japan Society for the Promotion of Science (Okuno, member), Grant-in Aid for Industrial Research from New Energy and Industrial Technology Development Organization (NEDO) (Takano, head), Grant-in-Aid for Young Scientists in Kyoto University (Takano, head), Core Research for Evolutionary Science and Technology (CREST) from Japan Science and Technology Agency (Takano, member)

### **A-4. International cooperations and overseas activities**

#### ***International meetings (roles)***

EMBO Workshop in Plant Virology, July 1-5 (2006), Helsinki, Finland, 1 oral presentation (Okuno)

25th Annual Meeting of American Society for Virology, July 15-19 (2006) Madison, WI, USA, 2 poster presentations (Okuno, Mise, Kaido)

Iwate Plant Science Symposium, October 18-19 (2006), Morioka, Iwate, Japan, 1 oral presentation (Takano)

24th Fungal Genetics Conference *Colletotrichum* Workshop, 3.25 (2007) Pacific Grove, CA, USA, 1 oral presentation (Takano)

#### ***Scholars from abroad***

“Shigaku Kenshu-in” trainee 1 (Lecturer, Shangqui Normal University, China)

### **B. Educational Activities (2006.4-2007.3)**

#### **B-1. On-campus teaching**

##### **a) Courses given**

Undergraduate level: Plant Pathology I (Okuno), Plant Pathology II (Mise, Takano), Seminar in Plant Protection (Okuno), Biotechnology-Novel Strategies for Agriculture (Okuno), Basic Bioresource Science II (Okuno), Microbiology (Okuno), Outline of Bioresource Science IV (Mise, Takano), Laboratory Course in Bioresource Science I, II (Okuno, Mise, Takano)

Graduate level: Plant Pathology (Advanced Course) I (Okuno), Seminar in Plant Pathology (Okuno, Mise, Takano), Research in Plant Pathology (Okuno, Mise, Takano)

#### **B-2. Off-campus teaching, etc.**

##### ***Part-time lecturer***

Mise, K.: Kyoto Institute of Technology (Plant Pathology)

### B-3. Overseas teaching

#### *Students and research fellows from abroad*

Master's Program: 1 (Thailand), 1 (China)

### C. Other remarks

Okuno, T.: The Safety Committee of Kyoto University for Recombinant DNA Experiments, The Steering Committee of Kyoto University Museum, A special research scientist in the Research Center for Science System in Japan Society for the Promotion of Science  
Mise, K.: The Committee of Graduate School of Agriculture for the Prevention of Radiation Injury  
Kaido, M.: The Committee of Graduate School of Agriculture for the Safety and Sanitation, The Committee of Kyoto University for Disposal of Inorganic Liquid Wastes

## 2.4.4 Laboratory of Insect Ecology

*Staff*      *Professor*                      : Fujisaki, Kenji, D. Agric. Sci.  
                 *Associate Professor* : Ohsaki, Naota, D. Agric. Sci.  
                 *Instructor*                        : Nishida, Takayoshi, D. Agric. Sci.

#### *Students and research fellows*

*Doctor's program*    : (17)  
*Master's Program*    : (9)  
*Undergraduate*       : (6)  
*Post doctoral fellow*: (8)

## A. Research Activities (2006.4-2007.3)

### A-1. Main subjects

The studies in this laboratory are concentrated on the ecology, dynamics, evolution and management of insect populations or communities with reference to agriculture and forestry. On-going main topics of research by the staff members are as follows.

- a) Analysis on life history strategy in insects - Insects have evolved various life history strategies in response to spatio-temporal heterogeneity of habitats. Among them, life history traits such as migration and diapause are especially important. These life history traits correlate with each other and constitute a syndrome. The evolutionary pathway of wing polymorphism as an example of such a syndrome is going to be clarified through the physiological, ecological and genetic analysis.
- b) Role of parasitic natural enemies in host plant choice by Pieris butterflies - Herbivorous insects do not always use nutritionally superior plants as food but use sometimes only inferior ones. This suggests the existence of other ecological factors influencing the food plant choice by herbivores. By studies of the host plant - herbivore - parasitoid interactions using Pieris butterflies, field and laboratory evidence is now being accumulated to infer that the difference in defense mechanisms of the insects against their specific parasitoids determines their host plant preference.

- c) In Batesian mimicry, mimetic forms of some species are female limited, other species are male limited, whereas the other species are not sex limited. To explain these phenomenon, trade-offs between predation by avian predators and sexual selection are analyzed.
- d) Indirect effect of predation on long-term population dynamics - Field observations and laboratory experiments have gradually revealed that prey often suffers more from indirect effects of predation (non-lethal injury, starvation, postponed reproduction and so on) than from direct predation events. This may explain why traditional analyses of population dynamics have long underestimated the effect of predation as the regulatory factor for prey-predator systems.
- e) Reproductive interference as a major underpinning mechanism of interspecific competition – Both theoretical and empirical studies have revealed that reproductive interference between closely related species can explain a wide spectrum of ecological phenomena, such as host range, habitat selection, geographic distribution, and biological invasion in herbivorous insects.
- f) Prediction of the expansion of distribution range and the population dynamics in insects as a biosensor of global warming – Climatic adaptation of some southern insect pests such as the cotton bollworm and the southern green stink bug to temperate climate zone has been analyzed. Based on the obtained data, the model of forecasting the seasonal occurrence under the future warming conditions is now constructing.
- g) Analysis of the host-egg parasite relationship in sting bugs-Researche on host-egg parasite relationship is not only important for the development of biological control but also interesting from the ecological aspect. The host-egg parasite relationship is now ongoing using eggs of some stink bugs and their parasitic wasps.

## A-2. Publications and presentations

### a) Publications

#### **Books**

- Ohsaki, N.: In search of mystery of Batian mimicry in Kakamega forest. Invitation to African Entomology (ed. T. Hidaka). P.13-31. Kyoto University Press, Kyoto, 2007 (in Japanese)
- Saulich A.H. and Musolin D.L.: *Seasonal Development of Aquatic and Semiaquatic True Bugs (Heteroptera)*. St. Petersburg University Press, St. Petersburg, Russia (203 pages), 2007 (in Russian, with an Extended English Summary)

#### **Original papers**

- Egusa, S., T. Nishida, K. Fujisaki, and H. Sawada: Spatio-temporal abundance of flushing leaves shapes host selection in the willow leaf beetle, *Plagioderma versicolora*. Entomologia Experimentalis et Applicata 120:229-237, 2006
- Himuro, C., T. Hosokawa, and N. Suzuki: Alternative mating strategy of small male *Megacopta punctatissima* (Hemiptera: Plataspidae) in the presence of large intraspecific males. Annals of the Entomological Society of America 99: 974-977, 2006
- Honma, A., S. Oku, and T. Nishida: Adaptive significance of death feigning posture as a specialized inducible defence against gape-limited predators. Proceedings of the Royal Society of London B- Biological Science 273: 1631-1636, 2006
- Ide, J.: Sexual and seasonal differences in the frequency of beak marks on the wings of two *Lethe* butterflies. Ecological Research 21: 453-459, 2006
- Iida, H. and K. Fujisaki: Seasonal changes in resource allocation within an individual offspring of the wolf spider, *Pardosa pseudoannulata* (Araneae: Lycosidae). Physiological Entomology

- Imai, K. and N. Ohsaki: Density-dependent egg mortality in early stages of gall induction by the fruit gall midge *Asphondylia aucubae* Yukawa et Ohsaki. *Ecological Research* 22:143-149, 2007
- Imai, K. and N. Ohsaki: Loss of integument in maturing fruits prevents gall induction by the midge, *Asphondylia aucubae* (Cecidomyiidae: Diptera). *Environmental Entomology* 35:1109-1114, 2006
- Kishi, M., T. Harada and K. Fujisaki: Dispersal and reproductive responses of the water strider, *Aquarius paludum* (Hemiptera: Gerridae), to changing NaCl concentrations. *European Journal of Entomology* (in press), 2007
- Kishi, S and T. Nishida: Adjustment of parental investment in the dung beetle *Onthophagus atripennis* (Col., Scarabaeidae). *Ethology* 112: 1239-1245, 2006
- Lopez Ruf, M; Perez Goodwyn, P.J.; Martins Neto, R.G.2006. New Heteroptera (Insecta) from the Santana Formation, Lower Cretaceous (Northeastern Brazil), with description of a new family and new taxa of Naucoridae and Gelastocoridae. *Gaea*. 1 (2): 68-74
- Miura, K. and N. Ohsaki: Examination of the food processes on mixed inferior host plants in a polyphagous grasshopper. *Population Ecology* 48: 239-243, 2006
- Musolin D.L. and Fujisaki K.: Changes in ranges: trends in distribution of true bugs (Heteroptera) under conditions of the current climate warming. *Russian Entomological Journal*. Vol. 15 (2): 175-179, 2006
- Musolin D.L., Fujisaki K. & Numata H.: Photoperiodic control of diapause termination, colour change and postdiapause reproduction in the southern green stink bug, *Nezara viridula*. *Physiological Entomology* 32:64-72, 2007
- Musolin D.L.: Insects in a warmer world: Ecological, physiological and life-history responses of Heteroptera to climate change. *Global Change Biology* Vol. 13, 2007 (in press)
- Perez-Goodwyn, P. J., A. Peressadko, H. Schwarz, V. Kastner and S. N. Gorb: Material structure, stiffness and adhesion: Why grasshoppers (*Tettigonia viridissima*) attach stronger than locusts (*Locusta migratoria*) (Insecta: Orthoptera)?. *Journal of Comparative Physiology A*. 192: 1233-1243, 2006
- Perez-Goodwyn, P. J., S. Ohba and J. A. Schnack: Chorion morphology of the eggs of *Lethocerus delpontei*, *Kirkaldyia deyrolli*, and *Horvathinia pelocoroides* (Heteroptera: Belostomatidae). *Rus. J. Entomol.* 15 (2) (I. Kerzhner Festschrift): 151-156, 2006
- Perez-Goodwyn, P. J. and K. Fujisaki (2006) Sexual conflicts, loss of flight, and fitness gains in locomotion of polymorphic water striders (Gerridae). *Entomologia Experimentalis et Applicata*. (accepted)
- Saulich, A. H. and Musolin, D. L.: Four seasons: Diversity of seasonal adaptations and ecological mechanisms controlling seasonal development in true bugs (Heteroptera) in the temperate climate. *Proceedings of the Biological Institute of St. Petersburg State University: A special issue in memory of Prof. Viktor P. Tyshchenko*. 53:25-106, 2007 (in Russian, with expanded 4-page English summary)(in press)
- Shimizu, K. and K. Fujisaki: Geographic variation in diapause induction under constant and changing conditions in *Helicoverpa armigera* (Hb.) (Lepidoptera: Noctuidae). *Entomologia Experimentalis et Applicata* 121: 253-260, 2006
- Tanaka, S. and N. Ohsaki.: Behavioral manipulation of hostcaterpillars by the primary parasitoid

- wasp *Cotesia glomerata* (L.) to construct defensive webs against hyperparasitism. Ecological Research 21:570-577, 2006
- Tanaka, S., Nishida, T. & Ohsaki, N: Sequential rapid adaptation of indigenous parasitoid wasps to the invasive butterfly *Pieris brassicae*. Evolution (in press)
- Tosaka, Y. and T. Nishida: Gall surface area as a simple and accurate measure of fitness in a galling aphid, *Neothoracaphis yanonis* (Homoptera: Aphididae). Applied Entomology and Zoology 42:217-222, 2007
- Yamazaki, K. and N. Ohsaki: Willow leaf traits affecting host use by the leaf-gall-forming sawfly. Population Ecology 48:363-371, 2006
- b) Conference and seminar papers presented
- The 50th Annual Meeting of Japanese Society of Applied Entomology and Zoology: 11 presentations
- The 53rd Annual Meeting of Ecological Society of Japan: 12 presentations
- Annual meeting of Society of Population Ecology: 1 presentation
- Annual meeting of Society of Animal Behavior: 3 presentations
- The third international symposium on Aero Aqua Bio-mechanisms ISABAMEC 2006. Ginowan, Okinawa: 1 presentation

### **A-3. Off-campus activities**

#### ***Membership in academic societies (roles)***

- Fujisaki, K.: The Japanese Society of Applied Entomology and Zoology (Executive Committee Member), The Society of Population Ecology (President, Editor)
- Ohsaki, N.: The Japanese Society of Ethology (Editor)
- Nishida, T.: Ecological Society of Japan (Editor), The Entomological Society of Japan (Editor), British Ecological Society (Associate Editor)

#### ***Research Grant***

- 21st century COE program: Innovative food and environmental studies pioneered by entomomimetic sciences (Fujisaki, K. project leader)
- JSPS grant for foreign researchers: Biomechanical studies on the locomotion systems of waterstriders on water surface
- Monbu-kagakusyo Research Grant: Scientific Research (C) (2) Effects of parasitoids on the evolution of host range of *Pieris* butterflies (Ohsaki, N.), Scientific Research: Nonlethal effects of predation on the community structure of grasshoppers in old fields (Nishida, T.), Scientific Research (C) Diversity and adaptive significance of domatia in *Viburnum* plants (Nishida, T.)

### **A-4. International cooperations and overseas activities**

#### ***International joint research, overseas research surveys***

- Fujisaki, K. Integrated biological control of *Pueraria thunbergiana* and *Polygonum perfoliatum*, invasive alien weeds in USA (USDA)
- Pablo Perez Goodwyn: Organizer of Japanese-German workshop on entomomimetics: taking ideas from insects. Max-Planck-Institute fuer Metallforschung, Stuttgart. 24 September 2006
- Pablo Perez Goodwyn: Invited scholar of Max-Planck-Institut fuer Metallforschung, Stuttgart. 1st - 20th November 2006

Pablo Perez Goodwyn: Invited Guest Researcher at Nanjing University of Aeronautics and Astronautics, Institute of Bio-Inspired Structure and Surface Engineering (IBSS)  
Duration 2 months (Dec. 2006-Feb. 2007)

***Scholars from abroad***

JSPS research fellow (Argentina)

COE foreign researcher (Russia)

**B. Educational Activities (2006.4-2007.3)**

**B-1. On-campus teaching**

a) Courses given

Undergraduate level: Insect ecology I (Fujisaki), Introduction to general bio-resource science II (Fujisaki), Biology of biosphere (Fujisaki), Insect ecology II (Ohsaki), Laboratory course in bio-resource science I and II (Ohsaki, Nishida), Seminar in plant protection (Fujisaki, Ohsaki)

Graduate level: Insect evolutionary ecology (Ohsaki), Seminar in insect ecology (Fujisaki, Ohsaki), Research in insect ecology (Fujisaki, Ohsaki)

**B. Educational Activities (2006.4-2007.3)**

**B-1. On-campus teaching**

a) Courses given

Undergraduate level: Outline of Bioscience III (Hirooka et al.), International Animal Agriculture (Ishida)

Graduate level: Seminar of Animal Husbandry Resources (Hirooka, Ishida), Laboratory Course of Animal Husbandry Resources (Hirooka, Ishida)

**B-3. Overseas teaching**

***Students and research fellows from abroad***

Master course student: 1 (Honduras)

## 2.4.5 Laboratory of Insect Physiology

*Staff      Professor                      : Sakuma, Masayuki, Dr. Agric. Sci.*

*Assistant Professor   : Fukui, Masao, Dr. Agric. Sci.*

*Students and research fellows*

*COE Postdoctoral Fellow: (1)*

*Doctor's program         : (5)*

*Master's program         : (2)*

*Undergraduate             : (1)*

### A. Research Activities (2006.4-2007.3)

#### A-1. Main subjects.

Notwithstanding their simple organization, the insects interact with environment by sophisticated behaviour. We are studying the unique mechanisms of sensory-motor system of insects and other arthropods, aiming for bio-mimetic engineering as well as insect pest control. Current topics of our research are as follows.

##### a) Study on the spatial orientation mechanisms of insects

We have been, and still are, developing locomotion-compensators (LC), such as a servo sphere LC, a micro LC, and a flight simulator, for precise analysis of sensori-motor context in insects and other arthropods. Since these apparatuses confine a test insect at one spatial position owing to a mechanical or visual feedback system, cues can be presented to the freely walking animal in a controlled way. This enables us to analyse the algorithms of the insect orientation behaviour.

The origin of chemo-orientation behaviour in arthropods could be found in the mould mite, *Tyrophagus putrescentiae*, living in complete still air conditions. The mite in the micro LC exhibited characteristic turns, which returns the mite to the odorous zone, only after the cessation of an attractant odour. The context of odour exposure leading to the strategic turn is under investigation.

We are developing a flight simulator for aviatory insects to survey sensori-motor contexts in flight. A tethered test insect is allowed to flap freely in the simulator, where a flowing pattern is displayed on a ground screen. Both the visual and olfactory cues steer the insect, so the simulator is now designed to control both cues to steer the insect autonomously. This year we developed a position sensor using a video-tracker for detecting the displacement of a tethered moth. The control system has been reviewed and replaced with a LabView OS.

##### b) Study on the odour discrimination mechanisms in insects

After convergence, insect's olfactory system represents great similarity to that of mammals. For the system analysis, however, it is advantageous to examine insect's neural network rather than mammals, because of far reduced number of nerve cells. Information processing time at a brain is important to know brain algorithms. We are measuring process time of odor discrimination and concentration tasks in an American cockroach brain. This year, we made a stimulator, which controls odorant concentration and stimulation timing with microsecond accuracy.



c) Odour learning in insects

In the insect olfactory behaviour, odour-learning is no less important than innate response to pheromonal odour. The odour-associative learning has been demonstrated in the German cockroach as the frequency of visits to the odour source. On a servosphere apparatus, however, the cockroaches exhibited different behavioural patterns to the general odour from those to a pheromonal odour.

d) Study on the semiochemicals of insects

The American cockroach is a well-known experimental animal as well as a notorious hygienic pest. Although its sex attractant pheromone was identified more than 30 years ago, an aggregation pheromone has not been identified yet. We continue purifying the aggregation attractant pheromones to isolate and identify the chemical, not only for the application purpose but also for use in research.

e) Study on the evolution of signals relevant to sexual selection

We examined effects of female preference on male song evolution in three *Teleogryllus* sibling species. A previous study of field crickets *Teleogryllus yezoemma* (*T<sub>y</sub>*), *T. emma* (*T<sub>e</sub>*), and *T. taiwanemma* (*T<sub>t</sub>*) suggested that calling songs of the species are effective for species recognition and pre-mating isolation. Males of two allopatric species (*T<sub>y</sub>*, *T<sub>t</sub>*) with similar songs were preferred by females of other species. A *T<sub>e</sub>* male song was discriminated by females of the two species. The *T<sub>y</sub>* female uses absolute measurement of the pulse interval duration to recognize male calls, whereas *T<sub>e</sub>* and *T<sub>t</sub>* females use the pulse rate. Crickets also use a courtship song when mating. Results of playback experiments with courtship songs showed that the *T<sub>y</sub>* female is attracted to the courtship songs of partially sympatric species *T<sub>e</sub>* and its allopatric species *T<sub>t</sub>*. These results suggest that the courtship song contributes little to species recognition at least by *T<sub>y</sub>*. These results indicated the close relation between *T<sub>y</sub>* and *T<sub>t</sub>*.

When on their host plants, male leafhopper *Nephotettix nigropictus* (Insecta; Hemiptera) produce substrate-borne signals to communicate with a female. After receiving a vibrational reply from a female responding to the calling signal, the calling male increases several components of the calling signal then continues in a duet with the female. The function of these signals, both influencing pair formation and the female's willingness to mate, is under study.

## A-2. Publications and presentations

a) Publications

**Original papers**

Fukui, M. (2006) Courtship stridulation by the male cockroach, *Nauphoeta cinerea* (Olivier) (Dictyoptera: Blaberidae) toward teneral adult and nymph. Jpn J Envir. Entomol. Zool. 17(3): 87-92

Honda-Sumi E., M. Sakuma, M. Fukui, T. Sekimoto and T. Hasegawa (2006) Oriented response to courtship song in the field cricket *Teleogryllus yezeomma*. The proceedings of the 9<sup>th</sup> Western Pacific Acoustic Conference (peer-reviewed) p.597-600

Kojima, T. and M. Sakuma (2006) Change in odour concentration steers walking mould mites. Proceedings of the 3<sup>rd</sup> International Symposium on Aero Aqua Bio-Mechanisms, (peer-reviewed) 6 pages in CD-ROM

b) Conference and seminar papers presented

2006 Annual Meeting of the Japan Society of Ethology, Okayama, October 2006: 1 poster

presentation

The 9<sup>th</sup> Western Pacific Acoustic Conference: 1 poster presentation

The 3<sup>rd</sup> International Symposium on Aero Aqua Bio-Mechanisms: 1 oral presentation

Japanese-German workshop on entomomimetics: taking ideas from insects: 2 presentations

The 4<sup>th</sup> Joint Meeting of the Acoustic Society of America and The Acoustic Society of Japan  
(Honolulu, Hawaii, USA): 1 presentation (Fukui, M. invited)

The 51<sup>st</sup> Annual Meeting of Japanese Society of Applied Entomology and Zoology: 4 presentations

### **A-3. Off-campus activities**

#### ***Research grants***

21<sup>st</sup> Century COE program: COE for Innovative Food and Environmental Studies Pioneered by  
Entomomimetic Sciences (Sakuma)

Sakuma, M.: Grant-in-Aid for Scientific Research from the Japan Society for the Promotion of  
Science: Grant-in-Aid for Scientific Research (C)(2) (Sakuma, head)

### **A-4. International cooperation and overseas activities**

#### ***International meetings***

Japanese-German workshop on entomomimetics (Sakuma, organizers member)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### **a) Courses given**

*Undergraduate level:* Animal Physiology (Sakuma et al.), Outline of Bioresource Science IV  
(Sakuma et al.), Insect Physiology (Sakuma), Laboratory in Bioscience I, II (Sakuma and  
Fukui), Seminar in Plant Protection (Sakuma), Introduction to Insect Behavioral  
Physiology (Sakuma)

*Graduate level:* Seminar in Insect Physiology (Sakuma), Research in Insect Physiology (Sakuma),  
Insect Physiology Advanced Course (Sakuma)

### **B-2. Off-campus teaching, etc.**

#### ***Part-time lecturer***

Sakuma, M. Faculty of Agriculture, Kobe University, (Integrated Pest Management Course II for  
JICA students)

Fukui, M. Faculty of Engineering, Kyoto Sangyo University, (Fundamentals of Biological  
Experiments)

# Chair of Animal Genetics and Reproduction

## 2.4.6 Laboratory of Animal Breeding and Genetics

*Staff Associate Professor: Yamada, Takahisa, Dr. Medic. Sci.*

*Assistant Professor : Taniguchi, Yukio*

*Assistant Professor : Miyake, Takeshi*

*Students and research fellows*

*Doctor's program : (2)*

*Master's program : (2)*

*Undergraduate : (1)*

### A. Research Activities (2006.4-2007.3)

#### A-1. Main subjects

##### a) Studies on Genetic Evaluation Procedure and Breeding Scheme for Beef Cattle

Heterogeneity of variance among subclasses of an effect is a potential source of bias in genetic evaluation. We quantified the heterogeneity of variance in carcass weight in Japanese Black cattle, and developed an adjustment method that was based on a maximum likelihood method with a log-linear model. As a result of adjustment, the heterogeneity of variance significantly reduced. The sire evaluation seemed to be robust against heterogeneity of variance. On the other hand, re-ranking of the elite dams due to the adjustment was observed. It is suggested that the impact of the adjustment on bull-dam selection is large. The effectiveness of the adjustment was evaluated in terms of the ability to predict breeding values. The results suggest that the genetic evaluation becomes more accurate by adjusting the data using the procedure developed in this study.

##### b) Studies on Analysis of Quantitative Trait Loci in livestock animals

As a strategy of locating Quantitative Trait Loci (OTL) on chromosomes, it is effective to use information on the linkage disequilibrium among genetic markers in the tagetted population of livestock. The multilocus association study was applied for the carcass weight and rib eye area of Japanese Brown cattle, and the suggestive regions for the location of QTLs were narrowed remarkably. Moreover, the MQEM program which was developed in our laboraroty for the multiple QTL and Epistasis mapping for outbred populations was revised with the Grid sampling approach and the executable speed of the program has been remarkably improved.

##### c) Exploration of the Genes Responsible for Beef Marbling

We are exploring beef marbling genes based on differential gene expression pattern between the two groups of the high-marbled Japanese Black and the low-marbled Holstein steers. We have suggested that +166 SNP in EDG1 promoter region is associated with marbling trait, and that G allele at the SNP has an effect on increase of beef marbling, by using Japanese Black sires and half-sib progeny of superior sires. Immunohistochemical analysis revealed that EDG1 is expressed in endothelial cells and adipocytes. Further, Reporter assay showed that EDG1 promoter activity is indistinguishable between G allele and A allele at +166 SNP in endothelial cells, suggesting that the SNP has no effect on transcriptional and translational efficiency of

EDG1. These results suggested that +166 SNP has an effect on EDG1 expression in endothelial cells and adipocytes and is associated with marbling trait, through molecular mechanism which is not detected by reporter assay, and/or that a SNP in linkage disequilibrium with +166 SNP does.

#### d) Study on the Molecular Mechanism Underlying Marbling in Beef

To establish the mouse model showing intramuscular fat deposition, transgenic mice expressing the bovine ADAM12 gene specifically in muscle tissue were produced and their phenotypes were analyzed. Transgenic mice did not differ from littermate controls in body weight and fat content. However, transgenic mice developed the increased intramuscular fat deposition more than littermate controls. Also, these results suggested that ADAM12 metalloprotease domain was sufficient to induce the intramuscular fat deposition.

## A-2. Publications and presentations

### a) Publications

#### *Original papers*

- Odani, M., A. Narita, T. Watanabe, K. Yokouchi, Y. Sugimoto, T. Fujita, T. Oguni, M. Matsumoto, and Y. Sasaki: Genome-wide linkage disequilibrium in two Japanese beef cattle breeds. *Anim. Genet.* 37; 139-144, 2006
- Yamada, T., Y. Taniguchi, S. Nishimura, Y. Yoshioka, A. Takasuga, Y. Sugimoto, and Y. Sasaki: Radiation hybrid mapping of bovine genes showing intramuscular fat deposition-associated expression changes in *Musculus longissimus* muscle. *Anim. Genet.* 37; 184-185, 2006
- Muramatsu, Y., H. Y. Lejukole, Y. Taniguchi, B. A. Konfortov, H. Yasue, T. Yamada and Y. Sasaki: Somatic cell hybrid mapping of expressed sequence tags for genes showing early embryonic death-associated changes of expression patterns in the fetal placenta of the cow carrying somatic nuclear-derived cloned embryo. *Anim. Biotechnol.* 18; 55-59, 2007
- Yamada, T., S. Ohtani, T. Sakurai, T. Tsuji, T. Kunieda and M. Yanagisawa: Reduced expression of endothelin receptor type B gene in piebald mice caused by an insertion of a retroposon-like element in intron 1. *J. Biol. Chem.* 281; 10799-10807, 2006
- Sasaki, Y., T. Yamada and Y. Sasaki: Quantitative and molecular genetic approaches for the improvement of carcass traits in the wagyu cattle. *Proceedings of the 8th World Congress on Genetics Applied to Livestock Production.* p21, 2006
- Miyake, T., A. Narita, M. Kurosawa, H. Hakaoka, T. Watanabe, K. Yokouchi, Y. Sugimoto, M. Ito, T. Fujita, T. Yamada and Y. Sasaki: Detection of epistatic QTLs for beef marbling by Bayesian MQEM method in Japanese Black cattle. *Proceedings of the 8th World Congress on Genetics Applied to Livestock Production.* p51, 2006
- Miyake, T., A. Narita, T. Yamada and Y. Sasaki: Bayesian multiple QTL mapping with epistasis for half-sib populations of beef cattle incorporating non-random mating under field records. *Proceedings of the 30th International Conference on Animal Genetics.* p23, 2006
- Matsumoto, K., H. Kose, S. Tsukumo, K. Yasutomo, T. Sakai, K. Wei and T. Yamada: Deletion in receptor-like protein tyrosine phosphatase kappa gene causes defective CD4<sup>+</sup> T cell development in the thymus in LEC rat. *Proceedings of the Australian Health and Medical Research Congress 2006.* p394, 2006

#### *Reviews*

- Miyake, T., T. Yamada, H. Nakaoka, A. Narita and Y. Sasaki: Useful properties of Bayesian

multiple QTL mapping with MCMC. *J. Anim. Genet.* 34: 17-29, 2006

Taniguchi, Y.: Future of studies on beef marbling: from identification of responsible genes to understanding of gene network. *J. Anim. Genet.* 34: 53-61, 2006

Sasaki, Y.: Transition of selection breeding systems in beef cattle: From external characteristics to gene. *Journal of System Research for Animal Husbandry.* 30: 3-12, 2006

Sasaki, Y.: Beef cattle breeding systems for improvement of beef production using field records. *Yougyu No Tomo. No.1-No.5 (July-November)*, 2006

### ***Reports***

The Development Group for the Beef Cattle Breeding System Based on Field Records (Chair: Y. Sasaki): Development of the genetic evaluation systems using field records and its utilizations (Highest award in the section of research and development, The 2005 annual grand prize to the best stock raisen). *Consultant for Animal Husbandry.* 42: 31-38, 2006

b) Conference and seminar papers presented

The 107th Annual Meeting of Japanese Society of Animal Science: 4 presentations

The 7th Annual Meeting of Japanese Society of Animal Breeding and Genetics: 4 presentations

## **A-3. Off-campus activities**

### ***Membership in academic societies***

Yamada, T: Japanese Society of Animal Breeding and Genetics (director)

### ***Research grants***

Monbusho research grant; Grant-in-Aid for Scientific Research (C) Establishment of mouse model showing intramuscular fat deposition by introducing the bovine *ADAM12* gene (Head: Taniguchi)

Collaboration research grant; Research grant in collaboration with BIG Research Institute, Exploration and identification of the genes responsible for beef marbling (Head: Yamada, Sharer: Taniguchi, Miyake). Research grant in collaboration with Livestock Improvement Association of Japan, INC. and Nippon Ham Co. Ltd., Analysis of the genes associated with beef marbling (Head: Yamada, Sharer: Taniguchi, Miyake)

## **A-4. International cooperations and overseas activities**

### ***Membership in international academic societies***

Sasaki, Y.: The 8th World Congress on Genetics Applied to Livestock Production. (Brazil)

Miyake, T.: The 8th World Congress on Genetics Applied to Livestock Production. (Brazil)

Miyake, T.: The 30th International Conference on Animal Genetics. (Brazil)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

a) Courses given

Undergraduate level: Outline of Bioresource Science II (Yamada et al.), Animal Breeding and Genetics (Sasaki), Introduction to Foreign Literature in Bioresource Science II (Yamada et al.), Basic Laboratory Course in Bioresource Science I, II (Yamada, Taniguchi, Miyake et al.), Technology theory of Animal Husbandry and Practice II (Yamada, Taniguchi, Miyake et al.), Laboratory Course in Bioresource Science I, II (Yamada, Taniguchi, Miyake et al.), Molecular Biology (Yamada), Seminar in Applied Animal Science I, II

(Yamada et al.), Domestic Animal Genome Science Biotechnology (Yamada et al.),  
Biometrics (Miyake et al.)

Graduate level: Philosophy of Applied Bioscience (Yamada et al.) Animal Breeding and Genetics  
(Yamada), Animal Genomics (Yamada), Seminar in Animal Breeding and Genetics  
(Yamada), Laboratory Course in Animal Breeding and Genetics (Yamada)

### **B-3. Overseas teaching**

#### ***Students and research fellows from abroad***

Doctor's program: 1 (Kyrgyz Republic)

### **C. Other remarks**

Yamada, T.: Committee for National Bio-Resource Project "Rat" in Graduate School of Medicine,  
Kyoto University (Member), Committee on Promotion and Support of Establishment and  
Maintenance for Bovine Superior Genetic Resource in Livestock Improvement  
Association of Japan, INC. (Member)

## **2.4.7 Laboratory of Reproductive Biology**

*Staff*      *Professor*                      : Imai, Hiroshi, Dr. Agric. Sci.  
                 *Associate Professor*: Yamada, Masayasu, Dr. Medic. Sci.  
                 *Assistant Professor* : Minami, Naojiro, Dr. Agric. Sci.

#### ***Students and research fellows***

*Doctor's program* : (4)              *Master's program* : (6)  
*Undergraduate*    : (4)              *Research student* : (1)

### **A. Research Activities (2006.4-2007.3)**

#### **A-1. Main subjects**

##### **a) Development and Differentiation in Mammalian Embryos**

The regulatory factors required for in vitro maturation, fertilization and development of eggs are studied. We found that IVM/IVF embryos require some unknown factors from follicle cells during maturation for their further development, so now we are focusing on elucidation of the molecular mechanisms of this intracellular communication between oocytes and follicle cells. In addition, genes and proteins that are involved in the preimplantation development and differentiation are also studied using mouse embryo culture system.

##### **b) Production of clone animals derived from somatic cells**

It has become possible to produce clone animals derived from differentiated somatic cells using nuclear transfer technique, however, it is still unknown how can differentiated cells acquire totipotency during development. In addition, the great majority of reconstituted embryos die after nuclear transfer without involvement of ontogenesis. In our laboratory, reprogramming mechanisms of differentiated cells and embryonic anomaly during embryogenesis are examined using cell and molecular biological techniques.

c) Establishment of germ-line stem cells and their use for producing transgenic animals

Development of techniques to establish embryonic and spermatic stem cells having pluripotency is undertaken. Reconstitution of embryos and production of transgenic animals using the stem cells are studied. We are intending to utilize embryonic stem cells to apply transgenic technique for improvement of livestock animals.

## A-2. Publications and presentations

### a) Publications

#### *Original papers*

- Cao, L., H. Shitara, T. Horii, Y. Nagao, H. Imai, K. Abe, T. Hara, J-I. Hayashi and H. Yonekawa: The mitochondrial bottleneck occurs without reduction of mtDNA content in female mouse germ cells. *Nature Genet.*, 39: 386-390 (2007)
- Suzuki, T., Minami, N., Kono, T. and Imai, H.: Zygotically activated genes are suppressed in mouse nuclear transferred embryos. *Cloning and Stem Cells* 8:295-304 (2006)
- Watanabe, T., Takeda, A., Tsukiyama, T., Mise, K., Okuno, T., Sasaki, H., Minami, N. and Imai, H.: Identification and characterization of two novel classes of small RNAs in the mouse germline: retrotransposon-derived siRNAs in oocytes and germline small RNAs in testes. *Genes & Dev.* 20:1732-1743 (2006)
- Ikeda S., Saeki K., Imai H. and Yamada M. Abilities of cumulus and granulosa cells to enhance the developmental competence of bovine oocytes during in vitro maturation period are promoted by midkine; a possible implication of its apoptosis suppressing effects. *Reproduction*, 132: 549-557 (2006)
- Ikeda, S., J. M. Prendes, C. Alonso-Montes, A. Rodrigues, C. Diez, M. Kitagawa, H. Imai and E. Gomez: Apoptosis-independent poor morphology of bovine embryos produced by multiple ovulation. *Reprod. Domestic Anim.*, 41: 383-385 (2006)
- Fumiiwa Y., Imai H., and Yamada M.: Nuclear localization of NF- $\kappa$ B in mouse embryos at the 1-cell and 2-cell stages, and effects of N-acetyl-L-cysteine and pyrrolidine dithiocarbamate on their development *in vitro*. *Jpn. J. Embryo Transfer* 28: 58-67 (in Japanese) (2006)

#### *Review*

- Imai, H.: The Advancement of Clone Technology in the Past 10 Years. *Obstetric and Gynecologic World*, 59: 219-225 (2007)
- Imai, H.: Transition of Animal Reproduction in Cattle and Advancement of Clone Technology. *J. Anim. Prod. System*, 30: 13-16 (2006)
- Imai, H.: Clone Technology- The Past 10 Years. *Animal Production Technology*, 9: 24-27 (2006)
- Minami, N. and Tsukamoto, S.: Role of oocyte-specific genes in the development of mammalian embryos. *Reprod. Med. Biol.* 5:175-182 (2006)
- Miyamoto, K. and H. Imai: Reprogramming of somatic cell in xenopus egg cell-free system. *Jpn. J. Embryo Transfer Soc.*, 28: 81-87 (in Japanese) (2006)
- Tsukamoto, S., Imaichi, H., Kimura, K., Imai, H. and Minami, N.: Method of cDNA library construction in bovine blastocyst. *Jpn. J. Embryo Transfer Soc.*, 28:118-123 (in Japanese) (2006)

### ***Patents***

Minami, N. Imai, H. and Usami, M.: Frosen semen straw with RFID. No. 2006-264064

b) Conference and seminar papers presented

The 99<sup>th</sup> Annual Meeting of Japanese Society of Animal Reproduction: 2 presentations

The 29<sup>th</sup> Annual Meeting of Japanese Society of Molecular Biology: 2 presentations

The 8<sup>th</sup> Annual Meeting of Japan RNA Society: 1 presentation

### **A-3. Off-campus activities**

#### ***Membership in academic societies***

Imai, H.: Japanese Society of Animal Reproduction (Director), Japan Embryo Transfer Society (Vice President, Director), Japan Society for Reproductive Medicin (Director), Japan Society of Fertilization and Implantation (Director), Japanese Society of Zootechnical Science (Councilor), Kansai Society of Zootechnical Science (Councilor), Japanese Society of Reproductive Endocrinology (Councilor)

Yamada, M.: Japan Society for Reproductive Medicin (Councilor), The Japanese Society of Animal Reproduction (Councilor), Japan Embryo Transfer Society (Officer),

Minami, N.: Japan Embryo Transfer Society (Officer), Japan Society for Reproductive Medicin (Officer), The Japanese Society of Animal Reproduction (Editorial Board)

#### ***Research grants***

Monbusho Research Grants, Grant-in-Aid for Scientific Research (B)(2): Functional analysis of a novel gene, Oogenesin, which localizes in nucleus at the time of zygotic gene activation (Head: Minami)

Research project for utilizing advanced technologies in agriculture, forestry and fisheries: Establishment of stable and efficient technology of the production of bovine somatic clones (Head: Imai, Sharer: Minami)

### **A-4. International cooperation and overseas activities**

#### ***International meetings (roles)***

39<sup>th</sup> Society for the Study of Reproduction, Omaha (1 presentation: Imai, Minami)

The 3<sup>rd</sup> Asian Reproductive Biotechnology Conference, Hanoi (3 presentations: Minami)

20<sup>th</sup> IUBMB International Congress of Biochemistry and Molecular Biology and 11<sup>th</sup> FAOBMB Congress (2 presentations: Imai and Minami)

Cold Spring Harbor Laboratory Meeting: Mouse Molecular Genetics, Cold Spring Harbor (1 presentation: Imai and Minami)

International Genomic Imprinting Workshop Tokyo (1 presentation: Imai and Minami)

The 2006 Seoul Forum on Production of Xenogenic-organs Seoul (Lecture: Minami)

33<sup>rd</sup> International Embryo Transfer Society, Kyoto (4 presentations: Imai, Yamada, Minami)

### **B. Educational Activities (2006.4-2007.3)**

#### **B-1. On-campus teaching**

a) Courses given

Undergraduate level: Outline of Bioresource Science III (Imai et al), Developmental and Reproductive Technology (Yamada), Introduction to Animal Science Literature I (Yamada and Moriya), Methods and Techniques in Animal Reproduction Experimentation (Imai,



Yamada, Minami), Biotechnology (Yamada et al), Reproductive Physiology (Minami)  
Graduate level: Advanced Course of Reproductive Physiology (Imai), Reproductive Physiology-Seminar (Imai, Yamada, Minami), Laboratory Course in Reproductive Physiology (Imai, Yamada, Minami), Genetic Engineering in Developmental Biology (Yamada)

## **B-2. Off-campus teaching, etc.**

### ***Part-time lecture***

National Livestock Breeding Center (Imai)

Osaka City University Medical School (Yamada)

### ***Open seminar, etc***

Imai, H.: Transition of Animal Reproduction in Cattle and Advancement of Clone Technology, The 33<sup>rd</sup> Symposium for the Society of Animal Production System (Lecturer), 2006

Imai, H.: Cell to Animal – The Application of Clone Technology. The Symposium in the Graduate School of Agriculture. (lecturer), 2006

Imai, H.: The Advancement and Application of Clone Technology. The Frontier Seminar for Animal Genetics. 2006

Imai, H.: Legacy from Dolly. NHK Siten/Ronten. 2007

Imai, H.: Hello Dolly. NHK Commentary. 2007

Yamada, M.: Development of in vitro culture technique of mammalian preimplantation embryos, The 12<sup>th</sup> Symposium for the Japanese Society of Clinical Embryologist. 2007

## **B-3. Overseas teaching**

### ***Lecture and seminars***

Students and research fellows from abroad

Graduate Student: 2 (Korea, India)

## **C-1. Other remarks**

Imai, H.: Committee of Research Investigation of, Agriculture, Forestry and Fisheries Technical Information Society, Committee of Kyoto University Livestock Farm, Assessment Committee of Project Research of Ministry of Agriculture, Forestry and Fisheries, Assessment Committee of Research Grant of Ministry of Agriculture, Forestry and Fisheries, Member of Administration Committee of Human Genome and Gene Analysis of Kyoto University

Yamada, M.: Guest Researcher of Gifu Prefectural Livestock Institute, Guest Researcher of Toyama Prefectural Agriculture Research Center

Minami, N.: Guest Researcher of National Institute of Radiological Sciences

## Chair of Animal Anatomy, Nutrition and Resources

### 2.4.8 Laboratory of Nutritional Science

*Staff      Professor                      : Yano, Hideo, Dr. Agric. Sci.*  
*Associate Professor: Matsui, Tohru, Dr. Agric. Sci.*  
*Assistant Professor : Kawachi, Hiroyuki, Dr. Eng.*

*Students and research fellows*

*Doctor's program : (3)*  
*Master's program : (8)*  
*Undergraduate : (4)*

#### A. Research Activities (2006.4-2007.3)

##### A-1. Main subjects

###### a) Mineral nutrition and metabolism

We have studied mineral metabolisms using stable isotopes as tracers in animals. We found that magnesium absorption from drinking water was affected by dietary phytate. We determined the true and apparent absorption of zinc in pigs. We examined the expression of Mg transporters such as TRMP6 and 7 in sheep digestive tract and clarified that the gene expression of these transporters was weak in the small intestine compared to the expression in the forestomach or large intestine. We also found that magnesium deficiency increased muscle degradation and thus muscle weight. In addition, we could show that magnesium deficiency increased the expression of UCP2 and 3 mRNAs in the skeletal muscle.

###### b) Adipocyte differentiation and function

We investigated the effect of isoflavones on differentiation of 3T3-L1 preadipocyte. Daidzein treatment during the early phase of differentiation induced the adipocyte differentiation markers, but equol and estrogen suppressed them. Estrogen treatment throughout the differentiation period repressed adipocyte differentiation, although daidzein and equol treatment stimulated it. Taken together, the present study provides the evidence that the effects of estrogen, daidzein and equol on adipogenesis are different respectively. Three bovine Ob-R isoform cDNAs were cloned and the sequence analyses revealed that these cDNAs were bovine Ob-R isoforms, i.e., the long form (Ob-Rb), the middle form (Ob-Ra) and the short form (Ob-Rc). The deduced amino acid sequences suggested that the isoforms were single transmembrane proteins, and differed in the C-terminal amino acid sequences. The amino acid sequence of these bovine Ob-R isoforms showed high identity compared with the corresponding mouse isoforms. Ob-R isoform mRNAs were distributed in a wide range of bovine tissues. Ob-Ra mRNA level was positively correlated with Ob-Rb mRNA level in perirenal, subcutaneous and intermuscular adipose tissues.

###### c) Vitamins

We determined plasma concentrations of vitamin K homologues in horse and found that plasma MK4 concentration was lower in horses than in other animals. The increasing ingestion of vitamin K<sub>1</sub> increased its plasma concentration but not MK4 concentration. We found that some MK analogues including MK7 was observed in cattle plasma but not in horse plasma except for

MK4 and suggested that MKs production by intestinal microbes can be negligible in horses.

We clarified that the plasma vitamin C concentration decreased with fattening in beef cattle, which was associated with the reduction of body weight gain during fattening.

## **A-2. Publications and presentations**

### **a) Publications**

#### ***Original papers***

Inoue, Y., Y. Asai, H. Ohmori, H. Fujii, T. Matsui<sup>1</sup> and H. Yano: Changes in serum biochemical markers of bone cell activity in growing Thoroughbred horses. *Asian-Australasian Journal of Animal Science* 19; 1632-1637. 2006

Mori, M., L. Padilla, T. Matsui, H. Yano, Y. Matsui and H. Yamada: Effects of vitamin C supplementation on plasma vitamin C level and fattening traits in Japanese Black cattle on a fattening farm. 81; 15-19. 2006 (in Japanese)

Hishiyama, N., H. Kayanuma, T. Matsui, H. Yano, T. Suganuma, M. Funaba and H. Fujise: Plasma concentration of vitamin C in dogs with a portosystemic shunt. *Canadian Journal Veterinary Research* 70; 305-307. 2006

Fujihara, T., M. N. Shem and T. Matsui: The effect of exogenous purine supply on the endogenous excretion of purine derivatives in the urine of growing lambs. *Animal Science Journal* 77; 582-586. 2006

Matsui, T., H. Kobayashi, S. Hirai, H. Kawachi and H. Yano: Magnesium deficiency stimulated mRNA expression of tumor necrosis factor- $\alpha$  in skeletal muscle of rats. *Nutrition Research* 27; 66-68. 2007

Hirai, S., H. Matsumoto, N. Hino, H. Kawachi, T. Matsui and H. Yano: Myostatin inhibits differentiation of bovine preadipocyte. *Domestic Animal Endocrinology* 32; 1-14. 2007

Fujihara, T., M. N. Shem and T. Matsui: Urinary excretion of purine derivatives and plasma allantoin level in sheep and goats during fasting. *Animal Science Journal* 78; 129-134. 2007

Chung, K. Y., D. K. Lunt, H. Kawachi, H. Yano and S. B. Smith: Lipogenesis and stearyl-CoA desaturase gene expression and enzyme activity in adipose tissue of short- and long-fed Angus and Wagyu steers fed corn- or hay-based diets. *Journal of Animal Science* 85; 380-387. 2007

#### ***Review***

Kawachi, H.: Micronutrients affecting adipogenesis in beef cattle. *Animal Science Journal* 77; 463-471. 2006

Matsui, T. Significance of magnesium in animals. In "New perspectives in magnesium research" (Y Nishizawa, H Mori and J Durlach eds.) Springer London, pp. 381-391. 2006

Kawachi, H., S. Hirai: Activin- an autocrine and paracrine factor affecting adipogenesis. *The Lipid* 17; 63-67. 2006 (in Japanese)

Hirai, S., H. Kawachi, H. Yano and T. Kawada: Molecular mechanisms of adipocyte differentiation- newly known functions of C/EBP  $\beta$  and  $\delta$ . *Saibou* 38; 219-223. 2006. (in Japanese)

### **b) Conference and seminar papers presented**

The 107th Meeting of Japanese Society of Animal Science: 3 presentations

The 56th Meeting of Kansai Society of Animal Science: 2 presentations

The Spring Meeting of Japanese Society for Animal Nutrition and Metabolism: 1 presentation

### **A-3. Off-campus activities**

#### ***Membership in academic societies***

Yano, H.: Japanese Trace Nutrients Research Society (President), Japanese Association of Pet Animal Nutrition (Vice President), Japanese Society for Magnesium Research (Director), Japanese Society of Nutrition and Food Science (Councilor), Japanese Society of Nutrition and Food Science-Kinki Section (Councilor), Kansai Society of Animal Science (Councilor), Japanese Society of Animal Nutrition and Metabolism (Councilor), Society of Beef Cattle Science (Councilor), Japanese Society of Veterinary Science (Councilor)

Matsui, T.: Japanese Trace Nutrients Research Society (Councilor), Japanese Society for Magnesium Research (Councilor, Editor), Japanese Society of Animal Nutrition and Metabolism (Editor), Japanese Association of Pet Animal Nutrition (Editor), Society of Beef Cattle Science (Secretary), Kansai Society of Animal Science (Councilor, Editor), Japanese Society of Nutrition and Food Science-Kinki Section (Councilor)

#### ***Membership in Science Council of Japan, etc.***

Yano, H.: Science Council of Japan (Member), Council of Animal Science (Chairman)

#### ***Research grants***

Monbukagakusho Research Grants: Scientific Research (B) The reduction of environmental loading by utilizing food residues as feedstuffs (Yano), Scientific Research (C) The evaluation of trace mineral bioavailability using stable isotope in animals (Matsui). Research Project for Utilizing Advanced Technologies in Agriculture, Forestry and Fisheries: Efficient culture of euglena and technology for feed production using the residue of methane fermentation (Yano), Research Project for Utilizing Advanced Technologies in Agriculture, Forestry and Fisheries: Development of palatable beef by integration of breeding information (Yano and Kawachi)

### **A-4. International cooperations and overseas activities**

#### ***International meetings (roles)***

Matsui, T.: 2006 The 11<sup>th</sup> International Magnesium Symposium (Presentation)

#### ***Membership in international academic societies***

Yano, H.: World Association for Animal Production (Director)

#### ***International joint researches, overseas research surveys***

Kawachi, H.: Difference of American Wagyu and Angus cattle (Texas A&M, USA)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### **a) Courses given**

Undergraduate level: Basic Bioresource Science I (Matsui et al), Basic Bioresource Science II (Yano et al), Outline of Bioresource Science II (Yano et al), Applied Animal Sciences (Yano et al), Animal Physiology (Matsui et al), Animal Nutrition (Matsui and Yano), Physiological Nutrition of Animals (Matsui Kawachi and Yano), Fundamentals for the Experiments for Bioresource Science (Matsui and Kawachi et al), Laboratory Course in Bioresource Science I and II (Matsui and Kawachi et al), Introduction to Foreign

Literature in Bioresource Science IV (Matsui et al), Seminar for Applied Animal Science I and II (Matsui et al)

Graduate level: Metabolism and its Regulation of Animals (Matsui), Seminar in Nutritional Science of Animals (Yano and Matsui), Laboratory Course in Nutritional Science of Animals (Yano and Matsui)

## **B-2. Off-campus teaching, etc.**

### ***Part-time lecturer***

Yano, H.: School of Environmental Science, The University of Shiga Prefecture (Environmentology for Animal Production)

Matsui, T.: School of Environmental Science, The University of Shiga Prefecture (Environmentology for Animal Production)

## **B-3. Overseas teaching**

### ***Students and research fellows from abroad***

Doctoral course: 3 students (Korea and Honduras)

## **C. Other remarks**

Yano, H.: Subcommittee of Agricultural Technology, Evaluation Committee of Incorporated Administrative Agency, The Ministry of Agriculture, Forestry and Fishery (Specialist). Committee on Preparation of Tables of Feed Composition for Animals, The Ministry of Agriculture, Forestry and Fishery (Member). Policymaking Advisory Board of Food, Agriculture and Agricultural Community, The Ministry of Agriculture, Forestry and Fishery (Temporal Member). Committee of Grant-in-Aid for Scientific Research, Japan Society for the Promotion of Science (Member). Advising Committee of Science and Technology of Gifu Prefecture (Member). Evaluation Committee of Research Institutes of Gifu Prefecture (Chairman)

Matsui, T.: Committee on Standard Methods for Feed Analyses, Fertilizer and Feed Inspection Services (Member). Subcommittee of Agricultural Industry, Evaluation Committee of Incorporated Administrative Agency, The Ministry of Agriculture, Forestry and Fishery (Specialist). Japanese Committee on ISO TC34/SC10, The Ministry of Agriculture, Forestry and Fishery (Specialist)

## 2.4.9 Laboratory of Animal Physiology and Functional Anatomy

*Staff*      *Professor*                               : Kume, Shin-ichi, Dr. Agric. Sci.  
              *Assistant Professor*               : Sugimoto, Miki, Dr. Agric. Sci.  
              *Assistant Professor*               : Ikeda, Shuntaro, Dr. Agric. Sci. (2006.9.1-)

### *Students and research fellows*

*Research fellows of the Japan Society for the Promotion of Science: (1)*  
*Doctor's Program: (2)*  
*(Including 1 of Research fellows of the Japan Society for the Promotion of Science)*  
*Master's Program: (3)*  
*Undergraduate : (3)*

## A. Research Activities (2006.4-2007.3)

### A-1. Main subjects

- a) Clarification of the effect of environmental pollution on physiology and reproductive functions of mammals.

Endocrine disrupters can bind to nuclear receptors including estrogen receptors and interfere with the reproductive functions in animals. We have been evaluating the effects of endocrine disrupters and other environmental load substances on the biological functions in mammals to develop a method that reduces their undesired effects. Effects of endocrine disrupters on nuclear receptors in bones, kidneys and small intestines are being examined in pregnant mice and domestic animals.

- b) Assessments of the effect of global warming on physiology and productivity of mammals.

Animals convert ingested feed into heat energy and maintain their body temperature by homeostasis, while they yield livestock products for human food. Global warming, however, impairs the physiology and productivity of mammals. Effects of heat stresses on metabolism at the organ- to whole-body levels are being examined using biochemical and pathological techniques. In addition, methods for effective utilization of energy and materials upon environmental changes are being developed.

- c) Functional morphology of mammalian reproduction and its regulation mechanism

Domestic animals with high productivity often have low reproductive ability. We are analyzing the factors involved in different processes in mammalian reproduction and are developing techniques for efficient utilization of germ cells and for avoidance of reproductive disorders.

### A-2. Publications and presentations

- a) Publications

#### ***Original papers***

Kume, S., K. Nagano and K. Kimura: Effects of galacto-oligosaccharides supplementation on health and mineral status in dairy calves. *Trace Nutrients Research* 23:22-27, 2006

Ikeda, S., K. Saeki, H. Imai and M. Yamada: Abilities of cumulus and granulosa cells to enhance the developmental competence of bovine oocytes during in vitro maturation period are promoted by midkine; A possible implication of its apoptosis suppressing effects.

Reproduction 132: 549-557, 2006

Ikeda, S., J.M. Prendes, C. Alonso-Montes, A. Rodríguez, C. Díez, M. Kitagawa, H. Imai and E. Gómez: Apoptosis-independent poor morphology of bovine embryos produced by multiple ovulation. *Reproduction in Domestic Animals* 41: 383-385, 2006

Rodríguez, A., C. Díez, S. Ikeda, L.J. Royo, J.N. Caamaño, C. Alonso-Montes, F. Goyache, I. Alvarez, N. Facal and E. Gómez: Retinoids during the in vitro transition from bovine morula to blastocyst. *Human Reproduction* 21: 2149-2157, 2006

Gómez, E., C. Díez, J.N. Caamaño, F. Goyache, L.J. Royo, A. Rodríguez, C. de Frutos, N. Facal and S. Ikeda: Nuclear receptors, small molecules and reproductive biotechnology. *AETE Newsletter* 25:11-13, 2006

Chan, M.P., Morisawa S., Nakayama A., Kawamoto Y., Sugimoto M. and Yoneda M.: A physiologically based pharmacokinetic model for endosulfan in the male Sprague-Dawley rats. *Environmental toxicology* 21:464-78, 2006

### ***Reviews***

Kume, S.: Methan production and excretion of nitrogen and mineral in dairy cows. *Chikusan system kennkyukaiho* 30:25-33, 2006

### ***Reports***

Kume, S.: Calf nutrition and growth. *Manual on optimal growth of Japanese Black calves*. 58-62, 2007

b) Conference and seminar papers presented

The 142th Annual Meeting of Japanese Society of Veterinary Science: 1 presentations

## **A-3. Off-campus activities**

### ***Membership in academic societies***

Kume, S.: Japanese Society of Equine Science (Editor), Japanese Society of Animal Nutrition and Metabolism (Councilor)

### ***Membership in Science Council of Japan etc.***

Kume, S.: Science Council of Japan (Associate member)

### ***Research grants***

Kume, S.: Grant-in-Aid for Scientific Research from the Japanese Society for Promotion of Science (B) (Kume Sharer)

Sugimoto, M.: Grant-in-Aid for Scientific Research from the Japanese Society for Promotion of Science (C) (Sugimoto Head)

## **A-4. International cooperation and overseas activities**

### ***International meetings***

Kume, S.: 12th Asian-Australasian Association of Animal Production Societies (AAAP) Animal Science Congress 2006, Busan (Presentation), 33rd Annual Conference of the International Embryo Transfer Society, Kyoto (Presentation)

Sugimoto, M.: 12th Asian-Australasian Association of Animal Production Societies (AAAP) Animal Science Congress 2006, Busan (Presentation), 33rd Annual Conference of the International Embryo Transfer Society, Kyoto (Presentation)

Ikeda, S.: 33rd Annual Conference of the International Embryo Transfer Society, Kyoto (Presentation)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### **a) Courses given**

Undergraduate level: Outline of Bioresource Science II (Kume et al.), Animal Physiology (Kume et al.), Fundamentals for the Experiments of Bioresource Sciences (Sugimoto, Ikeda et al.), Animal Physiology and Anatomy (Kume), Introduction to Foreign Literature in Bioresource Science II (Kume et al.), Laboratory Course in Bioresource Science I and II (Sugimoto, Ikeda et al.), Applied Animal Sciences (Kume et al.), Animal Environmental Physiology (Kume), Livestock Production Techniques and Practice II (Kume et al.), Seminar for Applied Animal Science I and II (Sugimoto et al.)

Graduate level: Animal Physiology and Anatomy (Kume), Seminar in Animal Physiology and Anatomy (Kume), Laboratory Course in Animal Physiology and Anatomy (Kume)

### **B-2. Off-campus teaching, etc.**

#### ***Open seminar, etc***

Kume, S.: Global warming and milk production. Symposium in Japanese Society of Animal Science (lecture) 2006

## **C. Other remarks**

Kume, S.: Subcommittee of Japanese Standard for Beef Cattle (member), Assessment Committee of Research Grant of Ministry of Agriculture, Forestry and Fisheries (member), Agricultural Establishment Research Committee of Kinki, Chugoku, Shikoku Region in National Agriculture and Bio-oriented Research organization (member), Planning Committee of Oita Dairy Industry (member), Feeding Committee of Japan Livestock Technology Association (member)

## **2.4.10 Laboratory of Animal Husbandry Resources**

*Staff            Professor                                : Hirooka, Hiroyuki, D. Agric. Sci.*

*Associate Professor: Kumagai, Hajime, D. Agric. Sci.*

*Assistant Professor : Oishi, Kazato*

*Students and research fellows*

*Doctor's program : (3)*

*Master's program : (6)*

*Undergraduate        : (2)*

## **A. Research Activities (2006.4-2007.3)**

### **A-1. Main subjects**

#### **a) Total evaluation for animal production systems.**

Data and information on genetic performance, nutrition, management and economic situations for beef, dairy, pigs, sheep and goats were collected from various research fields



concerning animal sciences in order to evaluate total animal production systems. Modeling and simulation of such systems were carried out.

b) Studies on livestock production systems in tropical areas.

Surveys have been conducted to investigate various performances of native livestock, nutrient requirements of the livestock, feeding system, quality of feeds and grassland production in tropical areas. Such information will be used to search better strategies for improving the existing systems. Sustainable farming systems integrated between livestock and crops in smallholders in the developing countries were evaluated through modeling works. The surveys have been conducted in the south area of Thailand and in the central area of Nepal.

c) Studies on regional properties of vegetation and nomadic livestock in arid areas.

For the purpose of defining and settling the problems found in traditional animal production systems in arid areas, the evaluations of regional woody and herbaceous plants as the feed resources with reference to the physiological properties of nomadic animals have been investigated. The studies were as follows. 1) Strategy of reclaiming the grassland devastated by over-grazing in Syria. 2) Feeding and management techniques of nomadic animals in Gobi district, Mongolia.

d) Studies on environmental problems in animal production.

Since animal industries have been specialized and scaled up, environmental pollution caused by such animal industries led to serious problems in society in Japan. Models for predicting excretion of nitrogen and phosphorus from animal production systems were developed and a country-wide fact-finding studies of the connection between crop and animal industries were performed in order to define the role of animal industry in establishing a sustainable agriculture.

f) Studies on effective utilization of unused resources as fodders of animals.

Use of by-products as feeds of ruminants have been considered as one of the ways to do resources recycling effectively. Therefore, evaluation of several by-products as feed resources, development of organic, chemical and physical treatments to improve forage quality of the by-products, and use of forages made from the by-products in dairy cattle and beef cattle productions have been carried out. The by-products included woody resources such as bamboos, residues of Yam production, fermented by-products, etc.

g) Other themes.

Studies on conservation of useful genes, evaluation of similarity of clones, ethical studies about advanced reproductive technologies, theoretical studies on system analysis and statistics, anthropological studies on ways of use of milk and meat, economical studies on consumption of animal products, setting of breeding objectives, etc.

## **A-2. Publications and presentations**

a) Publications

***Books***

Kumagai, H. Handbook for animal production (New edition). Chapter 5 and 6: Animal production in tropics and savannas. Edited by Ogimoto, K. Kodansha Scientific. Tokyo. 2006

***Original papers***

Tabata, Y., Ishida, J., Inamura, T., Noda, M. and Hirooka, H. 2006. Nitrogen and phosphorus utilization and their cycling in mixed farming systems for beef cow-calf and rice production. Bulletin of Beef Cattle Science, 82: 4-12. (In Japanese)

- Kumagai, H. and Ngampongsai, W. Comparative studies on dry matter intake, digestibility and nitrogen metabolism between Thai native (TN) and Anglo Nubian x TN bucks. *Small Ruminant Research*, 66:129-134. 2006
- T. Ibi., Kahi, A.K. and Hirooka, H. Effect of carcass trait fluctuations on genetic and economic evaluation of carcass traits in Japanese Black cattle. *Journal of Animal Science*, 84: 3204-3211. 2006
- Hirooka, H., Kahi, A.K. and van der Lende, T. A new mathematical method to predict protein and fat retention in domesticated mammals. *Proceedings of World Congress on Genetics applied to Livestock Production*. 13-18. Belo Horizonte. 2006
- Kahi, A. K., T. Oguni, Y. Sumio, and H. Hirooka. Direct and indirect predictions of the genetic merits in Japanese Brown cattle. *Proceedings of World Congress on Genetics applied to Livestock Production*. 3-48. Belo Horizonte. 2006
- Kahi, A.K., Rewe, T.O. and Hirooka, H. Research partnerships in relation to topics in animal breeding and genetics. *Journal of Animal Science*, 84: 2276-2283. 2006
- Choumei, Y., Terada, F. and Hirooka, H. Prediction and comparison of nitrogen excretions in dairy and beef cattle. *Nihon Chikusan Gakkaiho*, 77: 485-494. 2006. (Abstract in English)
- Tabata, Y., Kitagawa, M., Inamura, T., Ishida, J. and Hirooka, H. Farm level phosphorus utilization and cycling in the mixed farming system of beef fattening and rice production. *Nihon Chikusan Gakkaiho*, 77: 279-288. 2006. (Abstract in English)
- Rewe, T.O., Indetie, D., Ojango, J.M.K. and Kahi, A.K. Breeding objectives for the Boran breed in Kenya: Model development and application to pasture-based production systems, Online publication date: 30-Mar-2006
- Kahi, A.K. and Hirooka, H. Economic efficiency of Japanese Black cattle selection schemes utilizing crossbreeding with the Holstein breed. *Animal Science Journal*. 77:176-187. 2006
- Choumei, Y., Ibi, T. and Hirooka, H. Effects of attributes and consciousness of fattening farms households on carcass prices and traits. *Nihon Chikusan Gakkaiho*, 77: 289-294. 2006. (Abstract in English)
- Choumei, Y., Kahi A.K., and Hirooka, H. Fit of Wood's function to weekly records of milk yield, total digestible nutrient intake and body weight changes in early lactation of multiparous Holstein cows in Japan. *Livestock Production Science*, 104: 156-164. 2006
- Hirooka, H., Nomura, T., Sato, M., Muramoto, T., Yoneya, H. and Ozawa, S. Population structure and genetic diversity of indigenous beef cattle in Mishima island. *Journal for the society of indigenous animal breed in Japan*, 23: 173 – 188. 2006. (In Japanese)
- Hayashi, Y., Maharjan, K. L. and Kumagai, H. Feeding traits, nutritional status and milk production of dairy cattle and buffalo in small-scale farms in Terai, Nepal. *Asian-Australasian Journal of Animal Science*, 19: 189-197. 2006
- Mazumder, M. A. R and Kumagai, H. 2006. Analyses of factors affecting dry matter intake of lactating dairy cows. *Animal Science Journal*, 77: 53-62
- Choumei., Y., Terada, F. and Hirooka, H. Comparison of nitrogen utilization and excretion in dairy and beef cattle. *Proceedings of the 12th Animal Science Congress, The Asian Australasian Association of Animal Production Societies*, Busan, Korea. 2006
- Tabata, Y. and Hirooka, H. The effect of nitrogen and phosphorus recycles in mixed farming systems for rice production and beef fattening. *Proceedings of the 12th Animal Science*

Congress, The Asian Australasian Association of Animal Production Societies, Busan, Korea. 2006

Kikuhara, K. and Hirooka, H. Development and evaluation of a crop-livestock mixed farming model for dairy cattle in Japan. Proceedings of the 12th Animal Science Congress, The Asian Australasian Association of Animal Production Societies, Busan, Korea. 2006

Kumagai, H., Pradhan R., Hirata, K., Hayashi, Y. and Baba, E. 2006. Ruminal degradability and in vivo digestibility of ground bamboo. Proceedings of the 12th Animal Science Congress, The Asian Australasian Association of Animal Production Societies, Busan, Korea. 2006

### ***Reviews***

Tabata, Y. and Hirooka, H. Quantification of nutrient flows in animal husbandry and the case studies (1) – (3). Study of Animal Husbandry. 60(5): 539-544, 60(6): 669-672, 60(7): 790-800, 2006 (In Japanese)

Hirooka, H. How should the education about animal husbandry be in future ? Information of animal husbandry in Japan. Agriculture and Livestock Industries Corporation. 2006

b) Conference and seminar papers presented

The 107th Annual Meeting of Japanese Society of Animal Science: 4 presentations

The 44th Annual Meeting of Society of Beef Cattle Science: 2 presentation

The 4th Annual Meeting of Japanese Society of Goat Science: 1 presentation

## **A-3. Off-campus activities**

### ***Membership in academic societies***

Hirooka, H.: Japanese Society of Animal Science (Representative), Japanese Agricultural System Society (Director, Editor), Society of Beef Cattle Science (Secretary), Kansai Society of Animal Science (Councilor)

Kumagai, H.: Japanese Animal Production System Society (Secretary)

### ***Research grant***

Monbushou Research Grants: Grant-in-Aid for Scientific Research (B): Studies on development and evaluation of environmentally sound animal production systems aiming to crop and animal integration (Head: Hirooka), Grant-in-Aid for Scientific Research (C): Studies on effective utilization of unused mountainous forest by grazing cattles and goats (Head: Kumagai), Promotion Research: Evaluation of advanced Japanese beef cattle breeding schemes incorporating performance testing for blood serum insulin-like growth factor I and carcass traits (Head: Hirooka)

Other Research Grants: Research aid from National Agriculture and Food Research Organization: Optimization of beef cattle – whole crop rice mixed farming production system (Head: Hirooka), Research aid from Shinko Engineering Co., Ltd.: Studies on application of bamboos to feed resources of ruminants (Head: Kumagai), Research aid from Ajinomoto Co., Ltd.: Effects of nucleic acids on ruminal fermentation in ruminants (Head: Kumagai)

## **A-4. International cooperation and overseas activities**

### ***Scholars from abroad***

Inviting researcher: Doctor Kahi Alexander Kigunzu (Professor of Animal Husbandry at Egerton University)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### a) Courses given

Undergraduate level: Outline of Bioresource Science II (Hirooka et al.), Production of Animal Husbandry Resources (Hirooka), Fundamentals for the Experiments of Bioresource Science (Kumagai et al.), Introduction to Foreign Literature in Bioresource Science II (Kumagai et al.), Laboratory Course in Bioresource Science I, II (Kumagai et al.), Applied Animal Sciences (Kumagai et al.), Seminar in Applied Animal Science I, II (Kumagai et al.), Livestock Production Techniques and Practice II (Kumagai et al.)

Graduate level: Seminar of Animal Husbandry Resources (Hirooka, Kumagai), Laboratory Course of Animal Husbandry Resources (Hirooka, Kumagai), System Science for Animal Production (Hirooka)

### **B-3. Overseas teaching**

#### *Students and research fellows from abroad*

Doctor course student: 1 (Honduras)

Master course student: 1 (Zambia)

## **Chair of Marine Biological Resources**

### **2.4.11 Laboratory of Fisheries and Environmental Oceanography**

*Staff      Professor                      : Fujiwara, Tateki, Dr. Agric. Sci*

*Associate Professor: Kasai, Akihide, Dr. Agric. Sci.*

*Assistant Professor : Kobayashi, Shiho, MSc. Agric. Sci.*

*Students and research fellows*

*Doctor's program : (2)*

*Master's program : (3)*

*Undergraduate : (4)*

## **A. Research activities (2006.4-2007.3)**

### **A-1. Main subjects**

#### a) Analysis of the mechanism maintaining high productivity of coastal seas

Coastal seas are highly productive areas. To reveal mechanisms maintaining this high productivity and to find effective measures to sustain this productivity, we are studying nutrient dynamics in the Seto Inland Sea (SIS): Nutrient pools of the oceanic origin are located in the lower layer at the openings of SIS and that of terrestrial origin lies on the bottom of Harima-nada. These nutrients are transported to the central SIS by the estuarine circulations and then upwell into the upper layer at Bisan Strait (central SIS).

b) Study on eutrophication and hypoxia in semi-enclosed coastal seas

Excess amount of loads of nitrogen and phosphorus flowing into Ise Bay, Tokyo Bay and Seto Inland Sea causes eutrophication and hypoxia in these seas. Mechanisms generating hypoxic water mass were studied and modeled.

c) Analysis of long term variability in coastal water qualities

To restore water qualities in semi-enclosed coastal seas (Ise Bay, Tokyo Bay and Seto Inland Sea), reduction of anthropogenic loads of organic matter (COD), phosphorus and nitrogen have been conducted for more than 30 years. Owing to these measures, near-shore water quality has been significantly improved. However, water qualities in basin-wide scale (WQBS) are not so. We gathered data of water qualities, hydrographic parameters and meteorological parameters over 20 ~ 30 years to make data-base. It was revealed that WQBS varies with low correlation with the amount of pollutant loads, but varies in relation to hydrographic and meteorological conditions. In Seto Inland Sea, variations in WQBS are governed mainly by the through flow of SIS.

d) Variability of nutrients in coastal waters

Variability of nutrient in coastal waters is studied by nitrogen stable isotope analysis. Dissolved inorganic nitrogen in the lower layer in Ise Bay shows clear seasonal change, accompanied by water intrusion from Pacific Ocean. New ammonium is created in the lower layer in spring, when the bottom water is isolated. This ammonium changed to nitrate in early summer. Nitrite peak is also observed in summer, indicating denitrification. When the cooling effect destroys the bottom hypoxia in autumn, the pooled nitrogen is transported to the euphotic layer and is used by primary production.

e) Analysis of marine Ecosystem

Marine food chains are analyzed by stable isotope ratio of nitrogen and carbon in muscular tissue of marine animals and particulate organic matter. Depending on the habitat environments, some bivalves can digest terrestrial organic matter, while others mainly digest only phytoplankton or benthic micro algae which are produced in the sea. It is found that most animals in estuaries digest marine produced organic matter but cannot digest terrestrial matter. On the other hand, oysters show similar isotope trends to particulate organic matter in the sea, indicating it can digest terrestrial matter.

## A-2. Publications and presentations

a) Publications

***Original papers***

Tateki Fujiwara: Estuarine circulation in river-mouth areas and enclosed coastal seas. *Bulletin of Coastal Oceanography*, 44, 95-106, 2007

Masaki Nakajima and Tateki Fujiwara: Estuarine circulation and hypoxic water masses in Osaka Bay, *Bulletin of Coastal Oceanography*, 44, 157-163, 2007

Katsuyuki Abo, Satoru Toda and Tateki Fujiwara: Nutrients distribution and residual currents in the eastern Seto Inland Sea in winter. *Coastal Engineering*, 53, 2006

Maki Kunii and Tateki Fujiwara: Through flow of the Seto Inland Sea driven by the sea level difference between western and eastern boundaries. *Sea and Sky*, 81, 63 – 72, 2006

Tateki Fujiwara, Shiho Kobayashi, Maki Kunii, and Natsuko Uno: Nitrogen and phosphorous in Seto Inland Sea: their origin, budget and variability. *Bulletin of Coastal Oceanography*, 43, 129-136, 2006

- Toshinori Takashi, Tateki Fujiwara, Toshiaki Sumitomo and Wataru Sakamoto: Prediction of slope water intrusion into the Kii Channel. *J. Oceanography*, 62, 105-113, 2006
- Akihide Kasai, Tatsuo Yamada and Hiroshi Takeda: Flow structure and hypoxia in Hiuchi-Nada, Seto Inland Sea, Japan. *Estuarine, Coastal and Shelf Science*, 71: 210-217, 2007
- Akihide Kasai, Ryo Sugimoto and Satomi Akamine: Formation mechanism of subsurface chlorophyll maximum in a coastal embayment. *Sea and Sky*, 82: 53-60, 2007
- Shiho Kobayashi, Tateki Fujiwara, Mituo Tada, Hideshi Tsukamoto, Toshihiko Toyoda: The distributions of nitrogen, phosphate, silicon and nutrient ratio during the stratified season in the Seto Inland Sea. *Oceanography in Japan*, 15: 283-297, 2006
- Shiho Kobayashi, John H. Simpson, Tateki Fujiwara, Kevin J. Horsburgh: Tidal stirring and its impact on water column stability and property distributions in a semi-enclosed shelf sea (Seto Inland Sea, Japan). *Continental Shelf Research*, 26:1295-1306, 2006
- Shiho Kobayashi, Tateki Fujiwara: Seasonal variation in intrusion depths from straits to adjoining basins in the Seto Inland Sea. *Sea and Sky*, 82(1): 1-11, 2006
- Shiho Kobayashi, Tateki Fujiwara, Yoshio Takasugi: A mechanism of matter transport around tidal front in land-locked region. *Coastal engineering in Japan*, 53: 921-925, 2006
- Shiho Kobayashi, Tateki Fujiwara, Akira Harashima: Seasonal and inter-annual variation of dissolved inorganic nitrogen in the Seto Inland Sea. *Bulletin on coastal oceanography*, 44(2): 165-175, 2007

### **Reports**

- Tateki Fujiwara: Generation mechanism of hypoxic water mass in Ise Bay. *Kaiyo Monthly*, 439, 5-8, 2007
- Shigeho Kakehi and Tateki Fujiwara: Modeling and hindcast of hypoxia in Ise Bay. *Kaiyo Monthly*, 439, 15-21, 2007
- Ryo Sugimoto, Tateki Fujiwara and Akihide Kasai: Dynamics of particulate organic matter and nutrients in Ise Bay. *Kaiyo Monthly*, 439, 9-14, 2007
- Yoshihiko Sugimoto and Tateki Fujiwara: Oxygen deficiency and restoration of Ise Bay. *Kaiyo Monthly*, 439, 3-4, 2007
- Tateki Fujiwara: Material transport in estuary. 201-208, Relationship among Forest, Land and Sea. pp.364, Kyoto University Press. 2007
- Tateki Fujiwara and Maki Kunii: Increases of turbidity and COD in Bisan Seto Seat analyzed with stable isotope method. Scientific Forum of the Seto Inland Sea, No. 47, 53-57, 2006
- Tateki Fujiwara and Yuichi Hayami: Huge internal wave and “Kyucyo” in Lake Biwa. *Bulletin of Japan Fisheries Resource Conservation Association*, No. 497, 3-6, 2006
- Tateki Fujiwara: Changing ecosystem and fisheries in coastal seas. Bulletin of Japan Fisheries Resource Conservation Association, No. 487, 3-5, 2005
- Akihide Kasai and Kosei Komatsu: Modeling the transport and survival processes of eggs and larvae of Japanese jack mackerel (*Trachurus japonicus*). *GLOBEC International Newsletter*, 12(2), 30-32, 2006
- Kazufumi Takayanagi, Chiyuki Sassa, Kosei Komatsu, Kenji Morinaga, Yuichi Tsukamoto, Yuichi Hirota, Kou Nishiuchi, Hideaki Nakata, Yoshiro Watanabe, Reiji Masuda and Akihide Kasai: FRECS: Fluctuation of recruitment of fish eggs and larvae by changes of spawning grounds and transport pattern in the East China Sea. *GLOBEC International Newsletter*, 12(2), 46-47, 2006

Komatsu K and Kasai, A.: Numerical model of transport of fish eggs and larvae. Elucidation of variability in fish stocks and development of advanced prediction of variability; 40-41, 2006

b) Conference and seminar papers presented

2006 Annual meeting of the Japan. Soc. Fish. Sci.: 2 presentations

2006 Spring meeting of the Oceanographic Soc. Japan: 4 presentation

2006 Autumn meeting of the Oceanographic Soc. Japan: 3 presentations

2006 Autumn meeting of Kinki Branch of the Fisheries Soc. Japan: 1 presentation

2006 Annual meeting of the Japan. Soc. Fish. Oceanogr.: 3 presentations

Coastal and Shelf Seas -Present Understanding and Future Challenges: 3 presentations

Pioneering Studies of Young Scientists on Chemical Pollution and Environmental Changes (Matsuyama, Japan): 1 presentation

Regional symposium of the Japan. Soc. Fish. Oceanogr. : 1 presentation

Symposium in Kyushu-Okinawa: 1 presentation

North Pacific Marine Science Organization (PICES) 15th annual meeting (Yokohama): 1 presentation

2006 RIAM Symposium: 1 presentation

2006 Symposium of Iwaki River Research group: 1 presentation

2006 Annual meeting of JSCE Coastal Engineering Committee.: 2 presentations

### **A-3. Off-campus activities**

#### ***Membership in academic societies***

Fujiwara, T.: The Oceanographic Society of Japan (Counselor), Coastal Oceanography Division of the Oceanographic Society of Japan (Editor, Committeeman), The Marine Meteorological Society (Director, Editor), The Japanese Society of Fisheries Oceanography (Committeeman), Japanese Society of Civil Engineers (Committeeman)

Kasai, A.: Coastal Oceanography Division of the Oceanographic Society of Japan (Editor, Committeeman), The Japanese Society of Fisheries Oceanography (Committeeman)

#### ***Research grants***

Fisheries Agency; Variability of fisheries resources and its prediction (Kasai)

Monbusho Research Grant: Scientific Research (C) The effect of increase of jerry fish accompanied by hypoxia on the ecosystem in coastal waters (Kasai), Scientific Research (B) Behavior of terrestrial organic matter and its effect on the production in coastal seas (Kasai), Scientific Research (A) The origin and characteristics of the ecosystem in the Ariake Bay head (Kasai), Scientific Research (B) Physics and high primary production in mixed waters (Kasai)

### **A-4. International cooperation and overseas activities**

#### ***International meetings (roles)***

Kasai, A.: Pioneering Studies of Young Scientists on Chemical Pollution and Environmental Changes (Matsuyama, Japan, Oral presentation), North Pacific Marine Science Organization (PICES) 15th annual meeting (Yokohama, Japan, Poster presentation), Coastal and Shelf Seas -Present Understanding and Future Challenges (Bangor, UK, Oral presentation)

Kobayashi, S. and Fujiwara, T.: Coastal and Shelf Seas -Present Understanding and Future Challenges (Bangor, UK, Oral presentation), Workshop on Coastal Observatories - Best practice in the synthesis of long-term observations and models, Proudman Oceanographic Laboratory (Liverpool, UK, Oral presentation)

International Joint Researches, Overseas Research Surveys

Kasai, A.: Cooperative research on the physics and high production in Menai Strait (UK)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### a) Courses given

Undergraduate level: Marine Environment (Fujiwara), Marine Ecosystem (Kasai), Practical Course in Marine Bioscience and Technology (Fujiwara, Kasai), Laboratory Course in Bioresource Science (Kasai), Outline of Bioresource Science (Fujiwara), Seminar in Marine Bioresources Science (Fujiwara, Kasai)

Graduate level: Physical Oceanography (Fujiwara), Marine Ecosystem Dynamics (Kasai), Seminar in Fisheries Physics (Fujiwara, Kasai), Laboratory Course in Fisheries Physics (Fujiwara, Kasai)

### **B-2. Off-campus teaching, etc**

#### *Part-time lecturer*

Kasai: Kinki University, Faculty of Agriculture (Statistics)

## **C. Other Remarks**

Fujiwara: Researcher of the Maritime Safety Agency, Researcher of the Disaster Prevention Research Institute, Researcher of the Disaster Science Research Institute, Kyoto University, Science and Policy Commissioner of International EMECS Center

Kasai: Researcher of the center for science and technology trends, Researcher of the GLOBEC committee, Researcher of the biological purification committee

Kasai: Best paper award of Fisheries Science (2006)



## 2.4.12 Laboratory of Marine Stock-Enhancement Biology

*Staff Professor : Tanaka, Masaru, Dr. Agric. Sci.*

*(Nakabo, Tetsuji, Dr. Agric. Sci. Ichthyology, Kyoto University Museum)*

*Associate Professor: Tagawa, Masatomo, Dr. Sci.*

*Assistant Professor : Nakayama, Kouji, Dr. Agric. Sci.*

*Students and Research Fellows*

*Doctor's program : (3)*

*Master's program : (9)*

*Undergraduate : (4)*

*JSPS fellow : (2)*

### A. Research Activities (2006.4-2007.3)

#### A-1. Main subjects

##### a) Early life history of fishes

Occurrence, distribution, food habits and growth of larvae and juveniles in coastal marine fishes as flounder, sea bass, sea breams, tunas, and mackerels have been observed in the sea, being combined with physiological laboratory experiments concerning endocrine, osmoregulatory, and digestive functions. Recent particular focuses have been placed on the stock structure and impacts of mass-release of juvenile flounder on coastal ecosystem, and origin of the Ariake sea bass population related to its amphidromous life history.

##### b) Studies of systematic ichthyology

We are proceeding to the systematic study of marine fishes based on the specimens from the Indo-Pacific region. We have another project to study the taxonomy of some common coastal fishes cooperated with the study of early life histories of them (supervised by Prof. T. Nakabo, Kyoto University Museum).

##### c) Endocrinological study of fish development

Controlling mechanisms of fish metamorphosis, neoteny and migration are investigated, mainly during early life history using endocrinological methods. Involvements of maternal hormones (thyroid hormones and cortisol contained in unfertilized eggs) in early survival and osmoregulation before the onset of larval endocrine organ are examined in flounders, ice goby, and tilapia.

#### A-2. Publications and Presentations

##### a) Publications

###### **Books**

Tanaka, M.: How can link the early life history of Japanese flounder and forest-sato-sea system. In "New century of the Sea of Japan studies 6" (Gamou,T, and A.Takeuchi eds.). pp.256-265, Kadokawa Shyoten, 2006

Tanaka, M.: What is idea of forest-sato-sea linkage. In "Studies of forest-sato-sea linkage - in direction to integrated management from forest to sea" (Yamashita Y. ed). pp.309-336, Kyoto University Press, 2006

### *Original papers*

- Takeda Y. and M. Tanaka: Freshwater adaptation during larval, juvenile and immature periods of starry flounder *Platichthys stellatus*, stone flounder *Platichthys bicoloratus* and their reciprocal hybrids. J. Fish Biol. in press, 2006
- Alvarez M. D. C., R.P. Dominguez, and M. Tanaka: Digestive capacity, growth and social stress in newly-metamorphosed Japanese flounder. Env. Biol. Fish. in press, 2006
- Shoji J. and M. Tanaka: Effects of river flow on larval growth and survival of Japanese seaperch *Lateolabrax japonicus* (Pisces) in the Chikugo River estuary, upper Ariake Bay. J. Fish Biol. in press, 2006
- Shoji J. and M. Tanaka: Variability in recruitment of Japanese seaperch around the estuarine turbidity maximum zone of the Chikugo River estuary, upper Ariake Bay: effect of spring river flow. Scientia Marina in press, 2006
- Islam M. S. and M. Tanaka: Ontogenetic diet shift during larva-juvenile transformation in Japanese sea bass *Lateolabrax japonicus* in Ariake Bay, Japan. Marine Ecol. Prog. Ser. in press, 2006
- Yube Y., T. Iseki, M. Hibino, K. Mizuno, K. Nakayama, and M. Tanaka: Daily and food habits of *Lateolabrax latus* larvae and juveniles in the innermost shallow waters of Uwajima Bay, Japan. Fish. Sci. 72: 1236-1249, 2006
- Islam M. S., M. Hibino, M. Tanaka: Tidal and diel variations in larval fish abundance in an estuarine inlet in Ariake Bay: implication for selective tidal stream transport. Ecol. Res. 22: 165-171, 2006
- Makino H., R. Masuda, and M. Tanaka: Ontogenetic changes of learning capacity under reward conditioning in striped knifejaw. Fish. Sci. 72: 1117-1182, 2006
- Islam M. S. and M. Tanaka: Spatial and variability in nursery functions along a temperate estuarine gradient: role of detrital versus algal trophic pathways. Can. J. Fish. Aquat. Sci. 63: 1848-1864, 2006
- Islam M. S., M. Hibino, T. Ohta, K. Nakayama, and M. Tanaka: Long-term patterns in the diets of Japanese temperate bass *Lateolabrax japonicus* larvae and juveniles in Chikugo estuarine nursery ground in Ariake Bay, Japan. Estuaries and Coasts 29: 524-534, 2006
- Hibino M., T. Ohta, T. Isoda, K. Nakayama, and M. Tanaka: Diel and tidal changes in the distribution and feeding habits of Japanese temperate bass *Lateolabrax japonicus* juveniles in the surf zone of Ariake Bay. Ichthyol. Res. 53: 129-136, 2006
- Ueda H., H. Nagai, M. Hibino, and M. Tanaka: Redescription of a symbiotic poecilo-stomatoid copepod *Anthessius graciliunguis* Do & Kajiyhara from plankton: the second record of the species and first record to the male. Plankton and Benthos Research 1: 102-108, 2006
- Islam M. S., M. Hibino, and M. Tanaka: Distribution and diets of larval and juvenile fishes: influence of salinity gradient and turbidity maximum in a temperate estuary in upper Ariake Bay, Japan. Estuarine, Coastal and Shelf Science 68: 62-74, 2006
- Yoseda K., S. Dan, T. Sugaya, K. Yokogi, M. Tanaka, and S. Tawada: Effect of temperature and delayed initial feeding on the growth of malabar grouper (*Epinephelus malabaricus*) larvae. Aquaculture 256: 192-200, 2006
- Islam M. S., H. Ueda, and M. Tanaka: Spatial and seasonal variations in copepod communities related to turbidity maximum along the Chikugo estuarine gradient in the upper Ariake Bay, Japan. Estuarine, Coastal and Shelf science 68: 113-126, 2006

- Islam M. S., M. Hibino, T. Ohta, K. Nakayama, and M. Tanaka: Environmental effect on diet, fecundity and condition of an endangered fish *Neosalanx reganius* (Osmeriformes) in the Chikugo estuary, in the upper Ariake Bay, Japan. Aquatic Living Resources 19; 59-68, 2006
- Wada T., N. Mitsunaga, H. Suzuki, Y. Yamashita, and M. Tanaka: Growth and Habitat of spotted halibut *Verasper variegatus* in the shallow coastal nursery area, Shimabara Peninsula in Ariake Bay, Japan. Fish. Sci. 72; 603-611, 2006
- Tanaka Y., H. Yamaguchi, O. Tominaga, T. Tsusaki, and M. Tanaka: Relationships between release season and feeding performance of hatchery-reared Japanese flounder *Paralichthys olivaceus*: In situ release experiment in coastal area of Wakasa Bay, Sea of Japan. J. Exp. Mar. Biol. Ecol 330; 511-520, 2006
- Bolasina S., M. Tagawa, Y. Yamashita, and M. Tanaka: Effect of stocking density on growth, digestive enzyme activity and cortisol level in larvae and juveniles of Japanese flounder, *Paralichthys olivaceus*. Aquaculture 259; 432-443, 2006
- Kurihara T., M. Shikatani, K. Nakayama, and M. Nishida: Proximate mechanisms causing a morphological variation of a turban snail among different shores. Zool. Sci. 23; 999-1008, 2006
- Sharma J.G., W.-S. Gwak, R. Masuda, M. Tanaka, and R. Chakrabarti: Survival, growth and RNA/DNA ratio of *Pagrus major* cultured under three different feeding regimes during early development. Asian Fish. Sci. 19; 389-400, 2006
- Sharma J.G., R. Masuda, and M. Tanaka: Orientation behaviour of *Pagrus major* larvae exposed to UV-B radiation in laboratory conditions. Int. J. Rad. Biol. 83; 49-52, 2006
- Tanaka Y., T. Ohkawa, Y. Yamashita, and M. Tanaka: Geographical differences in stomach contents and feeding intensity of juvenile Japanese flounder *Paralichthys olivaceus*. Bull. Japanese Soc. Sci. Fish. 72; 50-57, 2006
- Shoji J., K.W. Suzuki, and M. Tanaka: Effect of tide and river flow on physical and biological properties in the estuarine turbidity maximum of the Chikugo River estuary during spring in 2005: evaluation as a nursery for the estuarine-dependent fish, Japanese sea perch *Lateolabrax japonicus*. Bull. Jpn. Soc. Fish. Oceanogr. 70; 31-38, 2006
- Tanaka M.: Relict estuarine ecosystem isolated from the continental coastal waters. Aquabiology 29; 3-9, 2007
- Suzuki K.W.: Migratory histories in Japanese temperate bass (*Lateolabrax japonicus*) juveniles from the Chikugo River estuary estimated by stable isotope method. Aquabiology 29; 40-46, 2007

### **Reports**

- Tanaka M.: Integration of Fisheries biology and conservation ecology associated with Ariake and Chinese temperate bass. Research report (2003-2005) of kaken-hi (basic research A), 0-273, 2006
- Nakayama K., T. Ohkawa, Y. Marukawa, Y. Tainosho, and M. Tanaka: Studies on genetic population structure and ecological-physiological characteristics of local populations of Japanese flounder (*Paralichthys olivaceus*). Bull. Fish. Res. Agency Suppl. 5 ; 139-142, 2006

b) Conference and seminar papers presented

The Sessile Organisms Symposium: 1

International Symposium on Bluefin Tuna: 1  
54th Annual Meeting of Ecological Society of Japan: 1  
H16 Larval Fish Meeting: 2  
H19 Annual Meeting of the Japanese Society of Fisheries Science: 12

### **A-3. Off-campus activities**

#### ***Membership in academic societies (roles)***

Tanaka: The Japanese Society of Fisheries Science (Vice president, Council member of Kinki Branch), Japan Aquaculture Society (Council member), The Japanese Society of Fisheries Oceanography (council member), The Ichthyological Society of Japan (council member)

Tagawa: The Japanese Society of Fisheries Science (committee of fisheries education, publication committee)

Nakayama: The Ichthyological Society of Japan (committee of information technology)

#### ***Research grants***

Monbusho Research Grant: Scientific Research (A); Origin and characteristics of “continental coast relict ecosystem” conserved in the end-most part of Ariake bay (Head: Tanaka, Collaborator: Nakayama), Scientific Research (C): Differentiation mechanisms of left-right asymmetry, and prevention of juvenile malformation during flatfish metamorphosis in aquaculture (Head: Tagawa), Research for the future; Wooden artificial reef-basic investigation to see a mechanism of ecological linking between forest and sea (Head: Tanaka), Scientific Research (B); Synthetic research on the function of professional knowledge and its education and cultivation (Collaborator: Tagawa), Scientific Research (S); Biological and biochemical researches on enhancement and culture for the bluefin tuna (Collaborator: Tanaka), Research for the future; Establishment of Oikopleura mass-culture and its effect on flounder development (Collaborator: Tagawa), Research grant for JSPS fellow; Early life history of spotted halibut for future resource recovery (Head: Wada), Research grant for JSPS fellow; “Continental coast relict ecosystem” in Ariake bay - a validation through river comparison and stable isotopes (Head: Suzuki)

Entrust Research Grant: 「Research on development of seed production and release techniques for establishing ecosystem-conservation culture system」 (Head: Tanaka, Collaborator: Nakayama)

### **A-4. International cooperations and overseas activities**

#### ***Membership in international academic society***

Tanaka, M.: Journal of Applied Ichthyology (editorial board), Aquatic Living Resources (Scientific Advisory Committee)

#### ***International Joint Researches, Overseas Research Surveys***

Tanaka, M.: Cooperative research on aquaculture and biology of a primitive heterosomata (Malaysia)

Tanaka, M., Nakayama, K.: Cooperative research on the sympatric distribution of spotted and non-spotted temperate bass in south-western Korea (Korea)

### ***Scholars from abroad***

Visiting scientist 1 (Associate Professor, University of Kyongsang)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### **a) Courses given**

Undergraduate level: Marine Ecology (Tanaka), Marine Physiology (Tagawa), Laboratory Course in Marine Resource Biology (Tagawa, Nakayama), Seminar in Fisheries Science (Tanaka, Tagawa, Nakayama), Course in Marine Bioscience and Technology II (Nakayama), Pocket Seminar (Tanaka, Tagawa, Nakayama), Animal Physiology (Tagawa), Introductory Laboratory Course in Bioresource Science (Nakayama), Overview of Bioresource Science III (Tanaka, Tagawa), Forest-Sato-Sea Linkage Science (Tanaka), Laboratory Course in Forest-Sato-Sea Linkage Science C (Nakayama), Introduction to Marine Biology (Tagawa), Pocket Seminar “Learn from Affan forest (CW Nicol)” (Tanaka), Pocket Seminar “Introduction to fish biology for fish lovers” (Tagawa, Nakayama), Pocket Seminar “Building wooden school house - restoration of wooden culture” (Tanaka), Pocket Seminar “field seminar - forest is a sweetheart of the sea” (Tanaka), Pocket Seminar “Nature in Niyodogawa basin, Kochi prefecture” (Tanaka)

Graduate level: Seminar in Marine Resource Biology (Tanaka, Tagawa, Nakayama), Laboratory Course in Marine Resource Biology (Tanaka, Tagawa, Nakayama), Biology of Marine Resources (Tanaka)

### **B-2. Off-campus teaching, etc.**

#### ***Part-time Lecturer***

Tanaka: Invited lectures in universities (4), high schools (3), and other organizations (6)

Tagawa: “Science detective lectures” in elementary school (1), and junior high school (2). Junior Campus 2007

## **C. Other remarks**

### ***Committees in the Faculty***

Tanaka, M.: Director of Field Education and Research Center, Council member, Curriculum committee, Council member of Education and Research, Council member of Graduate School of Global Environmental Studies, Council member of University Museum, Council member of Ecological Research Center, Kyoto University Evaluation Committee, Security Committee of University Information Technology, Working Group for Student Safety,

### ***Committees out of the University***

Tanaka, M.: Shiga Prefectural committee for adjustment of Lake-Biwa Fisheries, Evaluating committee member of National Research Institute of Aquaculture, Evaluating committee member of Agricultural Research of Shiga Prefecture

## Chair of Marine Microbiology

### 2.4.13 Laboratory of Physiology and Genetics of Marine Microbes

*Staff      Professor                      : Sako, Yoshihiko, Dr. Agric. Sci..*

*Assistant Professor : Yoshinaga, Ikuo, Dr. Agric. Sci.*

*Students and research fellows*

*Post-doctoral fellow : (1)*

*Doctor's program     : (2)*

*Master's program     : (9)*

*Research student     : (1)*

*Undergraduate        : (4)*

#### A. Research Activities (2006.4-2007.3)

##### A-1. Main subjects

In the sea, there are many different environmental conditions such as open ocean, deep sea, hydrothermal vents, polar circles, red tide sea, coastal area, where many kinds of marine microbes (bacteria, archaea, and microalgae) live. We study their diversity and strategies how to live and survive and their mutual relationships in such conditions. We, further, aim to develop the genetic resources from these marine microbes.

Our main themes are as follows:

- a) Studies on novel marine hyperthermophilic microorganisms and their development of genetic resources
  - (1) Physiological and ecological studies on thermophilic microorganisms thriving at marine hydrothermal environments
  - (2) Isolation and characterization of novel hyperthermophilic microbes from marine hydrothermal vents
  - (3) Search for the useful genes and enzymes from hyperthermophilic microbes
- b) Ecology of microbes in the marine environments
  - (1) Bioremediation for marine environments by using coastal marine bacteria
  - (2) Molecular ecological studies on novel denitrifying bacteria and in coastal area
- c) Ecological and molecular biological studies on the harmful algae
  - (1) Molecular ecological studies of toxic dinoflagellates
  - (2) Genomics of organelle DNA in toxic dinoflagellates

##### A-2. Publications and presentations

- a) Publications

###### ***Books***

Nishimura, H. and Sako Y.: Bio-hydrogen – Hydrogen production by bioprocess –“Front of new energy Driving for construction of environment harmonious energy system” Yoshikawa S.(ed.) Kagakudoujin p.115-120, Kyoto, 2006 (in Japanese)

Yoshinaga, Ikuo: Nitrogen cycle on the earth and microorganisms “What’s microorganisms –Let’s

know more about invisible lives around yourselves” Japanese Society of Microbial Ecology (ed) p.68-80, Tokyo, 2006 (in Japanese)

### **Original papers**

- Hosoi-Tanabe S. and Y. Sako: Development and application of fluorescence *in situ* hybridization (FISH) method for simple and rapid identification of the toxic dinoflagellates *Alexandrium tamarens* and *Alexandrium catenella* in cultured and natural seawater. Fish Sci 72; 77-82, 2006
- Tanaka R., S. Kawaichi, H. Nishimura and Y. Sako: *Thermaerobacter litoralis* sp. nov., a strictly aerobic and thermophilic bacterium isolated from a coastal hydrothermal field. Int J Syst Evol Microbiol 5; 1531-1534, 2006
- Hosoi-Tanabe S. and Y. Sako: Genetic differentiation in the marine dinoflagellates *Alexandrium tamarens* and *Alexandrium catenella* based on DNA-DNA hybridization. Plankton Benthos Res 1; 138-146, 2006
- Kamikawa R., J. Asai, Y. Miyahara, K. Murata, K. Oyama, S. Yoshimatsu, T. Yoshida and Y. Sako: Application of a real-time PCR assay to a comprehensive method of monitoring harmful algae. Microbes Environ 21; 163-173, 2006
- Hosoi-Tanabe S., I. Otake and Y. Sako: Phylogenetic analysis of noxious red tide flagellates *Chattonella antiqua*, *C. marina*, *C. ovata*, and *C. verruculosa* (Raphidophyceae) based on the rRNA gene family. Fish Sci 72; 1200-1208, 2006
- Nakagawa S., F. Inagaki, Y. Suzuki, B.O. Steinsbu, M.A. Lever, K. Takai, B. Engelen, Y. Sako, C.W. Wheat, K. Horikoshi and Integrated Ocean Drilling Program Expedition 301 Scientists: Microbial community in black rust exposed to hot ridge flank crustal fluids. Appl Environ Microbiol 72; 6789-6799, 2006
- Amano T, Yoshinaga. I, Okada K., Sako Y., Yamagishi T., and Suwa Y. Are anaerobic ammonium-oxidizing (anammox) bacteria responsible dinitrogen gas emission from coastal ecosystems of Japan?, In Frontiers Science Series, University Academy Press. Inc., Tokyo, Japan, 48: 417-420, 2006

### **Reviews**

- Kamikawa R., Tababe(Hosoi) S., Sako Y.: Development of molecular monitoring method of toxic and harmful algae BRAIN TECHNO NEWS 117: 28-31, 2006 (in Japanese)

### **Reports**

- Sako,Y.: Development of molecular detection and quantification method of paralytic shellfish poisoning causative planktons (in Japanese). Reports of counterplanning for protection of shellfish poisoning damage (Ministry of Agriculture), 2006. Development of molecular identification method of new red tide planktons (in Japanese). Reports of counterplanning for protection of red tide damage (Ministry of Agriculture), 2006

#### **b) Conference and seminar papers presented**

- The 2006 Annual Meeting of the Japanese Society of Fisheries Science: general presentation 5, symposium 1, mini-symposium 1
- The 2006 Kinki regional meeting of the Japanese Society of Fisheries Science: 1
- The 9<sup>th</sup> Meeting of the Japanese Society of Marine Biotechnology: 1
- The 22<sup>th</sup> Annual Meeting of the Japanese Society of Microbial Ecology: 5
- The 2006 Annual Meeting of the Japanese Society of Phycology: 1
- 16<sup>th</sup> Conference of International Society for Evolutionary Protistology: 1

12<sup>th</sup> International Conference on Harmful Algae: 2

3<sup>rd</sup> International Workshop on targeted HAB species in the East Asia Waters (EASTHAB 3): 1

11<sup>th</sup> International Symposium on Microbial Ecology: International Society for Microbial Ecology:  
1

International Symposium 2006 Pioneering Studies of Young Scientists on Chemical Pollution and  
Environmental Changes: 21<sup>st</sup> Century Program in Ehime University: 1

Okazaki Biology Conference, OBC 4 on Terra Microbiology 2: 1

### **A-3. Off-campus activities**

#### ***Membership in academic societies (roles)***

Sako, Yoshihiko: Japanese Society of Fisheries Science (Trustee, Chairman of Kinki Branch,  
Trustee in Kinki Branch, Chairman of Publication committee), Japanese Society of  
Microbial Ecology (Secretary), Japan Society for Archaea (Executive Committee Member),  
Japanese Society of Marine Biotechnology (Trustee)

Yoshinaga, Ikuo: Japanese Society of Microbial Ecology (Business affairs)

#### ***Research Grant***

Sako, Yoshihiko: Monbukagakusho Grants-in-Aid for Scientific Research (A), Massive collection of  
metagenomes occurring at deep-sea hydrothermal environments and their application to  
industrial processes (Head: Sako); Monbukagakusho Grants-in-Aid for Exploratory  
Research, Study on future hydrogenase applicable to hydrogen energy from marine  
hyperthermophilic archaea (Head: Sako), Monbukagakusho Grants-in-Aid for Scientific  
Research (B) (2), Structural biological studies of target DNA recognitions by  
endonucleases (Head: Keiko Kita [Div. of Applied Life Sciences], Collaborator: Sako),  
Entrust Research Grant by Ministry of Agriculture, Forestry and Fisheries: Technical  
development in counterplanning for protection of shellfish poisoning damage (Head:  
Sako), Technical development in counterplanning for protection of red tide damage  
(Head: Sako), COE for Microbial-Process Development Pioneering Future Production  
Systems (Head: Sakayu Shimizu [Div. of Applied Life Sciences], Collaborator: Sako)

Yoshinaga, Ikuo: Monbukagakusho Grants-in-Aid for Scientific Research (C), Development of  
supersensitive detection techniques for denitrification with using the molecular markers  
and mapping of denitrification hot spot around coastal area of Japan (Head: Yoshinaga);  
Monbukagakusho Grants-in-Aid for Scientific Research (B), Species composition analyses  
of algicidal bacteria around macroalgal beds and microalgal blooming, and the  
application of the algicidal bacteria against harmful red tides (Head: Ichiro Imai [Div. of  
Applied Biosciences], Collaborator: Yoshinaga); Monbukagakusho Grants-in-Aid for  
Scientific Research (B), Dynamics of organic land-source materials in coastal sea and  
estimation of their significance on biological production (Head: Yoh Yamashita [Field  
Science Education and Research Center], Collaborator: Yoshinaga); Entrust Research  
Grant by Ministry of the Environments, Molecular biological detection of microorganisms  
relating anaerobic ammonium oxidation in freshwater and seawater environments  
(Head: Yoshinaga)



## **A-4. International cooperations and overseas activities**

### ***Overseas research surveys***

Sako, Yoshihiko: Development of genetic diagnosis for toxic microalgae (USA, Korea), Search for extremophilic microorganisms at sub-seafloor biotopes in cooperation with IODP (USA)

### ***Editorial work for international journals (role)***

Yoshinaga, Ikuo: Exploration of anammox microorganisms in Thai and Vietnam

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### **a) Courses given**

Undergraduate level: Marine Microbiology I (Sako), Marine Microbiology II (Sako), Seminar of Marine Microbiology (Sako), Fundamentals for the experiments of Bioresource Science (Sako, Yoshinaga, Nomura), Basic Bioresource Science I (Sako), Biotechnology - New Strategies in Agriculture- (Sako), Basic Bioresource Science I (Sako, sharer), Microbiology (Sako)

Graduate level: Current Topics in Marine Microbiology (Sako), Seminar in Marine Microbiology (Sako), Laboratory Course in Marine Microbiology (Sako)

### **B-2. Off-campus teaching, etc.**

#### ***Part-time lecturer***

Sako, Yoshihiko: Nishinomiya High School (Specially Science Class)

#### ***Open classes***

## **C. Other Remarks**

### ***Committees***

#### ***Committees in the Faculty***

Sako, Yoshihiko: Member of Research Promotion Committee of Research activities

Yoshinaga, Ikuo: Health Supervisor

#### ***Committees out of the University***

Sako, Yoshihiko: Member of evaluation committee in Extremobiosphere Research Center of Japan Marine Science and Technology Center, Member of Research administrative committee of Geo Biotechnology Development Organization

Sako, Yoshihiko: Award of Fisheries Science in 2006

Sako, Yoshihiko: Poster Award of Microbial Ecology in 2006

Yoshinaga, Ikuo: Poster Award of Microbial Ecology in 2006

## 2.4.14 Laboratory of Marine Environmental Microbiology

Staff    Professor                    : Nakahara, Hiroyuki, Dr. Sci.  
Associate Professor : Imai, Ichiro, Dr. Agric. Sci  
Instructor                : Ajisaka, Tetsuro, Dr. Agric. Sci.

### Students and research fellows

Doctor's program : (4)  
Master's program : (8)  
Undergraduate    : (3)  
Research fellow    : (2)  
Research student : (1)

## A. Research Activities (2006.4 - 2007.3)

### A-1. Main subjects

#### a) Effects of eutrophication and loading of pollutants on fishery resources.

The exponential expansion of human development, industrialization and exploitation of various resources has resulted in an undesirable nutrient enrichment and contamination of aquatic environment. The loadings from municipal, industrial urban and agricultural pollutants are a serious threat to the conservation of the global aquatic ecosystem and fisheries resources. So, we are studying the effects of nutrient enrichment, heavy metals and agricultural chemicals on the aquatic primary producers, phytoplankton and benthic macroalgae, in laboratory and field conditions and in single species and community levels.

#### b) Ecophysiological studies on red tides and toxic algal blooms in coastal sea .

Red tide incidents with fishery damage and toxic algal blooms causing poisonings of bivalves have recently increased in frequency in the coastal seas of temperate and tropical areas. To understand the mechanisms of red tide and toxic algal bloom occurrences, the life history and physiological ecology of causative organisms are studied especially on toxic and fish-killing species. And further, algicidal bacteria are investigated for the trial for exterminating the red tides of harmful flagellates such as *Chattonella* spp. and *Heterosigma akashiwo* (Raphidophyceae). Diatom blooms occasionally cause fisheries damage to Nori culture in coastal sea such as Ariake Sea. The mechanisms for diatom bloom occurrences are investigated with reference to resting stage cell formation, vegetative growth and turbidity in the sea.

#### c) Taxonomical and ecological study of *Sargassum* (marine algae)

Populations variations of *Sargassum* plants from East Asia area (Japan, China and Korea) are studying from DNA analysis (co-operation with other scientists) and taxonomical/statistical analysis. Main target plants are *Sargassum patens*, *S. piluliferum*, *S. horneri* and *S. filicinum*, which are very common in the coasts. On the other hand, subgenus *Sargassum* those are growing in southern part of Japan, Southeastern Asia and Pacific countries, are studying by the taxonomical and ecological analysis.

#### d) Multilateral studies of freshwater green algae in the Mekong

Freshwater algae (Chlorophyta, Rhodophyta and Cyanophyta) have been used for food in the vicinity of the Mekong, South-east Asia. Multilateral studies on these algae is now studying in Laos.

## A-2. Publications and presentations

### a) Publications

#### **Books**

- Nakahara, H. and B. Murata (eds): "Subject-book of Marine Environmental Creation" 226p. NPO, Study Center of Osaka Bay, Osaka, 2007 (in Japanese)
- Edvardsen, B. and I. Imai: The ecology of harmful flagellates within Prymnesiophyceae and Raphidophyceae, *In Ecology of Harmful Algae* (ed. by E. Graneli and J.T. Turner), Ecological Studies Vol. 189, pp.67-79, Springer-Verlag, Berlin, 2006
- Salomon, P.S. and I. Imai I: Pathogens of harmful microalgae, *In Ecology of Harmful Algae* (ed. by E. Graneli and J.T. Turner), Ecological Studies Vol. 189, pp.271-282, Springer-Verlag, Berlin, 2006
- Imai, I. and M. Uchida: Microorganisms and Fisheries. In "What are microorganisms? Let's Learn More Immediate Life" (Japanese Society of Microbial Ecology eds.), pp.146-155. Nikkagiren, Tokyo, 2006 (in Japanese)
- Imai, I.: Phytoplankton. In "Water Environment Handbook" (Japan Society of Water Environment eds.), pp.420-423. Asakurashoten, Tokyo, 2006 (in Japanese)
- Imai, I.: Control of red tides. In "Subject-book of Marine Environmental Creation" (H. Nakahara and B. Murata eds.), pp.103. NPO, Study Center of Osaka Bay, Osaka, 2007 (in Japanese)
- Imai, I., Y. Fukuyo and S. Hiroishi (eds): "Advanced Researches on Shellfish Poisonings: Current Status and Overview" 149p. Koseishakoseikaku, Tokyo, 2007 (in Japanese)
- Ajisaka, T.: Cladophora, in T. Akimichi (ed.) "An Illustrated Eco-History of the Mekong Basin", Kobundo, Tokyo, Japan. pp. 44-45, 2007 (in Japanese)

#### **Original papers**

- Tsukada, H., S. Tsujimura and H. Nakahara: Seasonal succession of phytoplankton in Lake Yogo over 2 years: effect of artificial manipulation. *Limnology* 7(1); 3-14, 2006
- Fujita, Y. and H. Nakahara: Variations in the microalgal structure in paddy soil in Osaka, Japan: comparison between surface and subsurface soils. *Limnology* 7(2); 83-91, 2006
- Yamamoto, Y. and H. Nakahara: Importance of interspecific competition in the abundance of *Aphanizomenon flos-aquae* (Cyanophyceae). *Limnology* 7(3); 163-170, 2006
- Tsukada, H., S. Tsujimura and H. Nakahara: Effect of nutrient availability on the C, N, and P elemental ratios in the cyanobacterium *Microcystis aeruginosa*. *Limnology* 7(3); 185-192, 2006
- Imai, I., D. Fujimaru, T. Nishigaki, M. Kurosaki and H. Sugita: Algicidal bacteria isolated from the surface of seaweeds from the coast of Osaka Bay in the Seto Inland Sea, Japan. *Afr. J. Mar. Sci.* 28; 319-323, 2006
- Naito, K. and I. Imai I: Controlling iron availability to red tide causing phytoplankton. *Proc. Techno-Ocean 2006*, Paper No.18; 1-5, 2006
- Naito, K., M. Suzuki, S. Mito, H. Hasegawa, M. Matsui and I. Imai I: Effects of the substances secreted from *Closterium aciculare* (Charophyceae, Chlorophyta) on the growth of freshwater phytoplankton under iron-deficient conditions. *Plankton Benthos Res* 1; 191-199, 2006
- Shiraishi T, S. Hiroishi, K. Nagai, J. Go, T. Yamamoto and I. Imai: Seasonal distribution of the shellfish-killing dinoflagellate *Heterocapsa circularisquama* in Ago Bay monitored by an

indirect fluorescent antibody technique using monoclonal antibodies. *Plankton Benthos Res* 2; 49-62; 2007

Ajisaka, T.: Problems of the identification of the “*Sargassum duplicatum*” group. *Coastal Marine Research* 30(1); 174-178, 2006

Noiraksar, T., T. Ajisaka and C. Kaewsuralikhit: Species of *Sargassum* in the East coast of the Gulf of Thailand. *Science Asia* 32; 99-106, 2006

### **Reviews**

Imai, I., M. Yamaguchi and Y. Hori: Eutrophication and occurrences of harmful algal blooms in the Seto Inland Sea, Japan. *Plankton Benthos Res.* 1; 71-84, 2006

Imai, I.: Occurrence mechanisms and counter measures of shellfish poisonings in Japan. *Aquaculture Magazine* 43 (12); 21-23, 2006 (in Japanese)

Komatsu, T., Tatsukawa, K. and Ajisaka, T.: Studies on floating seaweeds: general view. *Kaiyo Monthly* 38(8): 543-546, 2006 (in Japanese)

Ajisaka, T.: On the taxonomical problem in *Sargassum duplicatum* Bory (*Sargassum*, Phaeophyceae) and its related species group, *Kaiyo Monthly* 38(8): 553-558, 2006 (in Japanese)

Ajisaka, T. and S. Uwai: Appearance of some special receptacles in *Sargassum horneri*/ *S. filicinum* group. *Kaiyo Monthly* 38(8): 553-558, 2006 (in Japanese)

Uwai, S., Komatsu, T., Tatsukawa, K., Kawai, H. and T. Ajisaka: Genetic comparisons among the drifting and local population of *Sargassum horneri*. *Kaiyo Monthly* 38(8): 563-569, 2006 (in Japanese)

Komatsu, T., Tatsukawa, K. and T. Ajisaka: Studies on drifting seaweeds: accompanying or attaching animals with the seaweeds and those in East China Sea. *Kaiyo Monthly* 38(11):757-760, 2006 (in Japanese)

Komatsu, T., Wang, W-D., Tatsukawa, K., Zhang, S-Y., Ajisaka, T., Uwai, S., Aoki, M., Tanaka, K. and T. Sugimoto: Distribution of *Sargassum horneri* along the Chinese coast, especially in Zhejiang Province. *Kaiyo Monthly* 38(11): 795-797, 2006 (in Japanese)

Komatsu, T., Mikami, A., Matsunaga, D., Sagawa, R., Ishida, K., Tatsukawa, K., Ajisaka, T., Tanaka, K., Aoki, M. and T. Sugimoto: Distribution of drifting seaweeds in East China Sea. *Kaiyo Monthly* 38(11): 807-810, 2006 (in Japanese)

### **Reports**

Imai, I., T. Ishida and T. Watanabe: Ecological characteristics of the distribution of diatom resting stage cells in sediments of the Ariake Sea. Report of Counterplanning for Fisheries Damages by Diatom Red Tides (Fisheries Agency); p.12-21, 2006 (in Japanese)

Imai, I., T. Masuyama and Y. Matsuyama: Search for microorganisms responsible for diarrhetic shellfish poisoning and development of feasible monitoring. Report of Counterplanning for Protection of Shellfish Poisoning Damage (Ministry of Agriculture, Forest, and Fisheries); p.1-16, 2006 (in Japanese)

Imai, I., A. Tanabe and K. Naito: Effects of macro- and micro-nutrients on the growth of diatoms. Report on Technical Developments for Reducing Fishery Damages of Nori Aquaculture by Diatom Red Tides, Upgrading Project of Agriculture, Forestry and Fisheries by Using of High Technology (Fisheries Agency); p.109-123, 2006 (in Japanese)

b) Conference and seminar papers presented

2006 Meeting of the Japanese Society of Scientific Fisheries: 4 presentations

2006 Meeting of the Japanese Society of Nutrition and Food Science: 1 presentation  
 Workshop of Recent Progress on the Research and Management of *Cochlodinium* Blooms: 1 presentation  
 12th International Conference on harmful Algae: 3 presentations  
 2006 Joint Meeting of the Plankton Society and the Benthos Society: 6 presentations  
 North Pacific Marine Science Organization 15th Annual Meeting: 4 presentations  
 3rd International Workshop on Targeted HAB Species in the East Asia Waters: 1 presentation  
 2006 Meeting of the Japanese Society of Fisheries Oceanography: 1 presentation  
 2006 Meeting of Kinki Branch of Japanese Society of Scientific Fisheries: 2 presentations  
 Asian GEOHAB (Global Ecology and Oceanography of Harmful Algal Blooms) Meeting: 1 presentation  
 The 31th Annual Meeting of Phycological Society of Japan: 4 presentations

### **A-3. Off-campus activities**

#### ***Membership in academic societies***

Nakahara, Hiroyuki: The Japanese Society of Scientific Fisheries (Council Member of Kinki Branch)  
 Imai, Ichiro: The Japanese Society of Scientific Fisheries (Council Member, Chairman of Committee of Protecting Fisheries Environment, Convener of the Symposium “Problems of Toxic Blooms in Japan”, Member of the Executive Committee of 5th World Fisheries Congress, Council Member of Kinki Branch), The Plankton Society of Japan (Executive Editor of English Journal, Plankton & Benthos Research, Member of Selection Committee of Best Paper Award), The Japanese Society of Phycology (Council Member, Member of Editorial Board), The Japanese Society of Fisheries Oceanography (Council Member)  
 Ajisaka, Tetsuro: Japanese Society of Phycology (Council Member)

#### ***Research grant***

Nakahara, Hiroyuki: Monbukagakusho Grant-in-Aid for Scientific Research (B) (2), Comparative analysis of algicidal bacteria in red tide areas and seaweed beds, and application of algicidal bacteria in seaweed beds to prevention of harmful red tides (Head: Ichiro Imai, Collaborator: Nakahara)  
 Imai, Ichiro: Monbukagakusho Grant-in-Aid for Scientific Research (B) (2), Comparative analysis of algicidal bacteria in red tide areas and seaweed beds, and application of algicidal bacteria in seaweed beds to prevention of harmful red tides (Head: Imai), Monbukagakusho Grant-in-Aid for Exploratory Research, Studies on development of mass culture of appendicularians and effects of appendicularians on the growth and developments of the larvae of Japanese flounder (Head: Yo Yamashita [Field Science Education and Research Center], Collaborator: Imai), Entrust Research Fund by Ministry of Agriculture, Forestry and Fisheries: Urgent counterplanning for red tides (Head: Imai), Upgrading project of Agriculture, Forestry and Fisheries by using of high technology (Head: Imai), Subsidy Grant Project of Fisheries Research Agency, Studies on mechanisms, forecasting and countermeasures for harmful red tides of *Cochlodinium polykrikoides* (Head: Mineo Yamaguchi [National Research Institute of Inland Sea], Collaborator: Imai)

Ajisaka, Tetsuro: Monbukagakusho Overseas Research (A), Distribution and ecological study of *Sargassum* beds in China and its contribution for drifting seaweeds (Collaborator: Ajisaka)

## **A-4. International cooperation and overseas activities**

### ***International meetings***

Imai, Ichiro: Workshop of Recent Progress on the Research and Management of *Cochlodinium* Blooms, Cheju, Korea (invited speaker); 12th International Conference on harmful Algae, Copenhagen (poster); North Pacific Marine Science Organization 15th Annual Meeting, Yokohama (symposium organizer, invited speaker); 3rd International Workshop on Targeted HAB Species in the East Asia Waters, Nagasaki (Oral presentation); Asian GEOHAB (Global Ecology and Oceanography of Harmful Algal Blooms) Meeting, Tokyo (invited speaker)

### ***Membership in international academic society***

Imai, Ichiro: North Pacific Marine Science Organization (Delegate of Japanese committee members in Harmful Algal Bloom section)

### ***International Joint Researches, Overseas Research Surveys***

Nakahara, Hiroyuki: Recent Changes in the Ecosystems of the Northern Coast and the Syr-darya Delta of the Small Aral Sea (Republic of Kazakhstan)

Imai, Ichiro: Prevention of harmful red tides of the dinoflagellate *Cochlodinium polykrikoides* (Korea)

Ajisaka, Tetsuro: Biodiversity in Southeast Asian coastal area (Indonesia), A Trans-Disciplinary Study on the Regional Eco-History in Tropical Monsoon Asia (Laos, Thailand)

## **B. Educational Activities (2006.4 - 2007.3)**

### **B-1. On-campus teaching**

#### **a) Courses given**

Undergraduate level: Aquatic Environmental Microbiology (Nakahara), Aquatic Microbial Ecology (Imai), Outline of Bioresource Science II (Nakahara Partaker), Basic Bioresource Science II (Imai, Partaker), Importance of Global Environmental Study (Imai, Partaker), Practical Course in Marine Bioscience and Technology (Nakahara, Imai, partaker), Seminar in Marine Microbiology (Nakahara, Imai, partaker), Laboratory Course in Bioresource Science I, II (Imai, Ajisaka, partaker), Fundamentals for Laboratory Course in Bioresource Science (Imai, Ajisaka, partaker), Marine Practice I, II (Imai, Ajisaka, partaker), Marine Practice III (Faculty of Science) (Ajisaka, partaker), Water world Biology (Open seminar) (Ajisaka, partaker)

Graduate level: Aquatic Environmental Microbiology (Nakahara), Aquatic Microbial Ecology (Imai), Seminar of Aquatic Environmental Microbiology (Nakahara, Imai and Ajisaka), Laboratory Course of Aquatic Environmental Microbiology (Nakahara, Imai)

Graduate School of Global Environmental Studies: Management of Global Resources and Ecosystems (Nakahara, partaker), Conservation of Coastal Ecosystems (Nakahara, Imai), Seminar in Environmental Management (Summer Practice) (Nakahara, Imai, Ajisaka, partaker)

## **B-2. Off-campus teaching, etc.**

### ***Part-time lecturer***

Nakahara, Hiroyuki: Coastal Ecosystems and Global Environment, Face-to-face instruction, The University of the Air; Project of Linkage between High School and Kyoto University, Bioscience C Course

Ajisaka, Tetsuro: Mariculture of seaweeds, Faculty of Agriculture, Kinki University; Marine Practice, Faculty of Science, Nara Women's University

### ***Open seminar***

Imai, Ichiro: Workshop on technical managements of aquaculture concerning harmful algal blooms and countermeasures, Kyoto Prefecture

## **B-3. Overseas teaching**

Ajisaka, T.: National Agriculture and Forestry Research (NAFRI) in Laos (Lecture)

### ***Students and research fellows from abroad***

Doctor Course Student 1 (Korea), Research fellow 1 (China)

## **C. Other remarks**

Nakahara, H.: Kyoto Prefectural committee for adjustment of inland fisheries; Temporary member of the committee of the Council of Environment, Ministry of Environment,

Imai, I.: North Pacific Marine Science Organization (Delegate of Japanese Committee Member of Harmful Algal Bloom section); Fisheries Agency of Japan (Committee member: Training course of harmful algal blooms, Counter measures for harmful algal blooms); Ministry of Environments (Committee member: Advisory committee of harmful algal blooms, Advisory panel of a project of Global Environment Research Fund), Guest Investigator of Ehime University (Center for Marine Environmental Studies), Guest Scientist of Kyoto University (Center for Ecological Research)

## Chair of Marine Bio-production

### 2.4.15 Laboratory of Technology of Marine Bio-products

*Staff Professor : Hirata, Takashi, Dr.Agric.Sci.*  
*Associate Professor: Sugawara, Tatsuya, Dr.Agric.Sci.*  
*Assistant Professor : Kinoshita, Masato, Dr.Agric.Sci.*

*Number of Students and research fellows*

*Doctor's program : (2)*  
*Master's program : (13)*  
*Undergraduate : (2)*  
*Special research student : (1)*

#### A. Research Activities (2006.4-2007.3)

##### A-1. Main subjects

a) Studies on functional components extracted from oceanic lives

Oceanic life has adapted itself to the specific environment different from land. For this adaptation, various bio-active substance are incorporated into or biosynthesized in their bodies. The objectives of this work are to screen these components and to elucidate their functions for human use. The progress hitherto made is summarized as follows; (1) Pigment-proteins from *Porphyra* sp. and carotenoids from brown algae and Dinoflagellates inhibit a degranulation of mast cells. We found that these carotenoids inhibit antigen-induced aggregation of the IgE receptor. These results suggests that these pigments have anti-inflammatory activities. (2) Carotenoids from algae were studied from the perspective of their absorption, metabolism through intestinal tract, and anti-proliferative activities of the metabolites against various cell lines. We found for the first time that esterification of oxygenated carotenoids by intestinal cells.

b) Studies on regulation of lipid metabolism via nuclear receptors and nuclear transcription factor.

Modulation of lipid metabolism is important to decrease the risk of life-style related diseases. Recently, it has been known the mechanism of lipid metabolic gene expression through nuclear receptors. In this study, we examined the effect of structural modified EPA (trans-isomers and oxidized forms) on lipogenic gene expression via liver-X receptor (LXR) and sterol regulatory element binding protein-1c (SREBP-1c) in HepG2 human hepatoma cells. Our results show that trans-isomers of EPA decrease the expression of lipogenic gene more potently the lipogenic gene expression than its intact form. We found that the hypolipidemic effect of TEPA can be attributed to a decrease not only in SREBP-1 but also in PGC-1b expression.

c) Studies on post-harvest development of melanogenesis cascade in crustaceans.

Invertebrates including crustaceans lack acquired immunity system. And a phenoloxidase system, which is non-inducibly present, is one candidate for recognition of foreign materials. However, this cascade system is often triggered during post-mortem storage, resulting in black spot development and seriously reducing the market quality of crustaceans. So far we confirmed that hemocyanin-derived phenoloxidase activity plays the major role in the black spot



development, especially in frozen-thawed prawns. This year we found that phenoloxidase activity of hemocyanin in kuruma prawn is effectively suppressed by carbon dioxide.

d) Production of transgenic fish and regulation of expression of transgene.

Transgenic technique is one of the most powerful tools for the investigation of gene function. In this study, we used medaka as a model. I optimized the gene transfer conditions using microinjection method. And I also developed some vector cassettes, which regulate tissue specific gene expression (in muscle, nerve system, and liver). I also adopt Cre-LoxP system and showed the utility of this system in medaka transgenesis.

e) Development of transgenic medaka for the detection of endocrine disrupting substances.

We started to produce an unique transgenic medaka strain which tells the existence of endocrine disrupting substances by green body color.

## A-2. Publications and presentations

a) Publications

### ***Books and Review***

Sugawara, T.: Studies on intestinal absorption and nutritional functions of glycolipids. J. Jpn. Soc. Nutr. Food Sci. 60: 11-17, 2007 (in Japanese)

### ***Original papers***

Sugawara, T., Y. Komura, H. Hagino, T. Hirata: Phycoerythrin contributes to the photo-oxidation of eicosapentaenoic acid in *Porphyra yezoensis* during light-exposure, *Journal of Food Science* 71: S486-S491, 2006

Zaima, N., T. Sugawara, D. Goto, T. Hirata: Trans geometric isomers of eicosapentaenoic acid lower cellular triacylglycerol mediated through sterol regulatory element binding protein-1c and peroxisome-proliferator-activated receptor- $\gamma$  co-activator 1 $\beta$ . *Journal of Lipid Research* 47: 2712-2717, 2006

Sugawara, T., N. Zaima, A. Yamamoto, R. Noguchi, S. Sakai, T. Hirata: Isolation of sphingoid bases of sea cucumber cerebroside and their cytotoxicity against human colon cancer cells, *Bioscience Biotechnology Biochemistry* 70: 2906-2912, 2006

Sugawara, T., K. Matsubara, R. Akagi, M. Mori, T. Hirata: Antiangiogenic activity of brown algae fucoxanthin and its deacetylated product, fucoxanthinol. *Journal of Agricultural and Food Chemistry* 54: 9805-9810, 2006

Fu, X., C. Xue, B. Miao, Z. Li, X. Gao, and T. Hirata: Distribution and seasonal activity variation of the proteases in digestive tract of sea cucumber *Stichopus japonicus*, *Fisheries Science*, 72(5), 1130-1132, 2006

Matsuda, M., Shinomiya, A., Kinoshita, M., Suzuki, A., Kobayashi, T., Paul-Prasanth, B., Lau, E., Hamaguchi, S., Sakaizumi, M., Nagahama, Y. DMY gene induces male development in genetically female (XX) medaka fish. *Proc Natl Acad Sci U S A*, 104, 3865-3870 (2007)

Aizawa, K., Yori, K., Kaminaga, C., Yamashita, T., Kinoshita, M., Oda, S., Mitani, H. Response of embryonic germ cells of the radiation-sensitive medaka mutant to  $\gamma$ -irradiation. *J. Radiat. Res.*, 48, 121-128 (2007)

Hano, T., Oshima, Y., Kinoshita, M., Tanaka, M., Mishima, N., Ohyama, T., Yanagawa, T., Wakamatsu, T., Ozato, K., Honjyo, T. Quantitative bioimaging analysis of gonads in olvas-GFP/ST-II YI medaka (transgenic *Oryzias latipes*) exposed to ethinylestradiol. *Environ. Sci. Technol.* 41, 1473-1479 (2007)

b) Conference and seminar presented

The 60th Annual Scientific Meeting of Japanese Society of Nutrition and Food Science: 2 presentations

The 14th Sendai Conference of Lipid Peroxide biology and Medicine: 1 presentation

The 45th Annual Meeting of the Japan Oil Chemists' Society: 1 presentation

The 53th Annual Meeting of the Japanese Society for Food and Technology: 3 presentations

Kinki branch meeting of the Society of Scientific Fisheries: 3 presentations

Annual Meeting of the Japan Society for Bioscience, Biotechnology, and Agrochemistry, 2007: 3 presentations

Annual meeting of the Society of Scientific Fisheries: 2 symposium

Annual Meeting of the Japanese Society of Packaging Science and Technology

Annual Meeting of the Society of Scientific Fisheries: 3 presentations

The 7th Korea-Japan, Japan-Korea Joint Symposium on Aquaculture

### **A-3. Off-campus activities**

#### ***Membership in academic societies (role)***

Hirata, Takashi: The Japanese Society of Fisheries Science (Symposium planning committee, Fisheries science education committee), World fisheries congress (Program committee), The Society of Packaging Science and Technology (Director, Editor), The Japanese Society for Food Science and Technology (Director of Kansai Branch, Councilor, Auditor)

Sugawara Tatsuya: The Conference on Lipid Peroxide Biology and Medicine (Executive Committee Member), The Japanese Society of Fisheries Science (Secretary of Kinki branch)

#### ***Research grants***

Monbusho Research Grant: Scientific Research; Studies on the novel pathway of melanogenesis in crustaceans and its postharvest control (Hirata, Sugawara, Kinoshita), Germination research; Allergic control by nuclear receptor PPAR gamma ligands derived from marine products (Hirata, Sugawara), Scientific Research; Visualization of water pollutants using transgenic medaka (Kinoshita), Creative Scientific Research; Establishment of Neuroglycobiology (Glycobiological approach for Neuroscience) (Kinoshita), Grant-in-Aid for Scientific Research on Priority Areas 'Mechanisms of Sex Differentiation'; Initiation of oogenesis in medaka (Kinoshita), Ministry of the Environment: ExTEND 2005 Basic study; Application of transgenic medaka for the assessment of endocrine disruptors on gonadal cells and the study of their recovery mechanism (Kinoshita), Young Research; Studies on the Lipid-lowering effect of oxidized poly-unsaturated fatty acids (Sugawara)

### **A-4. International cooperations and overseas activities**

#### ***International meetings (roles)***

Kinoshita, M.: The 6th Coordinator's meeting of JSPS Multilateral Core University Program in Ha Long City, Vietnam (Oral Presentation)

Kinoshita, M.: The 2006 POME Workshop in Ha Long City, Vietnam (Oral Presentation)

#### ***International Activity***

Hirata, T: Ocean University of China (Invited lecture)

Kinoshita, M: University of Putra, Malaysia (Lecture and Sampling)

## **B. Educational Activities (2006.4-2007.3)**

### **B-1. On-campus teaching**

#### a) Courses given

Undergraduate level: Outline of Bioresource Science II (Hirata), Fundamentals for the experiments of Bioresource Science (Sugawara), Technology of Marine Bioproducts I (Hirata), Technology of Marine Bioproducts II (Hirata), Introduction to Foreign Literature in Bioresource Science III (Hirata), Seminar in Marine Bio-production (Hirata, Sugawara), Laboratory in Bioresource Science I, II (Hirata, Sugawara, Kinoshita), Practical Course in Marine Bioscience and Technology III (Hirata, Kinoshita), Technology of Marine Biological System (Sugawara)

Graduate level: Laboratory Course in Technology of Aquatic Biological Products (Hirata, Sugawara), Seminar in Technology of Aquatic Biological Products (Hirata, Sugawara), Technology of Aquatic Biological Products (Hirata), Biochemistry of Aquatic Animals (Sugawara)

### **B-2. Off-campus teaching, etc.**

#### *Part-time lecturer*

Hirata, Takashi: Fukui Prefectural University

Kinoshita, Masato: Kyoto Prefectural University of Medicine

#### *Open seminar*

Hirata, Takashi: Packaging manager course (Lecturer)

Kinoshtia, Masato: Lecture for high-school students (Lecturer)

## **C. Other Remarks**

Hirata, Takashi: Kinki Agri.High Technol.( Director )

Sugawara, Tatsuya: Young Investigator Award of the 2006's JSNFS

## **2.4.16 Laboratory of Marine Biological Function**

*Staff Associate Professor : Toyohara, Haruhiko, Dr. Agric. Sci.*

*Students and research fellows*

*Doctor's program : (3)*

*Master's program : (7)*

*Undergraduate : (4)*

## **A. Research Activities (2006.4-2007.3)**

### **A-1. Main subjects**

#### a) Production of a marine extract library and development of its application

Due to the difficulties in the collection and species identification, marine organisms remain undeveloped as seeds for pharmaceutical use. We are now attempting to establish a chemical compounds library of marine organisms that have been collected by scuba diving or with

cooperation of fishermen and marine stations for the purpose of screening of pharmaceutically active compounds useful for human health. We collected more than 500 species of marine organisms including mainly algae and invertebrates. After alcohol extraction and following hexane, ethylacetate, n-butanol and water, we screened the anti-allergy activity by using extracts. We successfully identified the anti-allergy substances, enzyme inhibitors and antagonists through the collaborations with companies.

b) Ecological and biochemical studies on benthos in estuaries and coastal area

Estuaries and coastal areas play important roles in degrading man-made and natural substances eluted from rivers. Marine organisms mainly benthos living these areas are possibly involved in the degrading process, but the biochemical mechanism of it still remains unknown. Particularly, cellulose is the most abundant biomass on the earth and is suggested to contribute to the carbon circulation at the global level. We found that meiobenthos including small animals less than 1mm in the mud play an important role in the breakdown of cellulose.

c) Studies on the biomineralization of marine organisms

The shell is made of calcium carbonate and a small amount of proteins that give a variety of structural properties specific for each species including pearls. We recently found that spider silk proteins play important roles in the biomineralization process of shell formation. We searched the genes specifically expressed in the edge of oyster mantle to identify the gene involved in the biomineralization process and isolate two genes that showed the homology with spider silk proteins. This year, we successfully produced the artificial shell in vitro by using peptides designed based on the amino acid sequence of oyster spider silk protein.

d) Development of the genetically engineered transgenic cultured fish

Fish culture is very important to maintain the food production in Japan. Breeding by crossing, however, is very limited in case of fish culture compared with agriculture. We focused on red seabream, one of the most important cultured fish, and made a specific vector for red seabream carrying actin promoter. This year, we successfully made an efficient expression vector for red seabream.

## A-2. Publications and presentations

a) Publications

***Original papers***

1. K. Touhata, Y. Touhata Tokuda, M. Yamashita, H. Toyohara: Molecular cloning and characterization of a tissue inhibitor of metalloproteinase-2 from red seabream cultured cells Ken Fish. Sci., 2006; 72: 699-701
2. Y. Yokoyama, H. Hashimoto, S. Kubota, A. Kuriyama, Y. Ogura, S. Mizuta, R. Yoshinaka, H. Toyohara. cDNA cloning of Japanese oyster stress protein homologous to the mammalian 78-kDa glucose regulated protein and its induction by heatshock. Fish. Sci, 2006; 72: 402-209
3. A. Kasai, H. Toyohara, A. Nakata, T. Miura and N. Azuma. Food sources for the bivalve *Corbicula japonica* in the foremost fishing lakes estimated from stable isotope analysis. Fish. Sci., 2006 72:105-114

***Reports***

Toyohara, H: 2006 Annual report on the Bio-Design Project by the Ministry of Agriculture, Forestry and Fisheries, Japan. 2004 Annual report to Nippon Suisan Fund

b) Conference and seminar papers presented

The 2006 Annual Meeting of the Japanese Society of Fisheries Science (4)

The 2006 Annual Meeting of the Japanese Association of Bentholog (2)

The 2006 Annual Meeting of Marine Biotechnology Conference (2)

The 2006 Aquagenome conference (1)

The 2006 Aquagenome symposium (1)

The 2006 Aquagenome symposium Biwako (1)

The 7<sup>th</sup> Japan-Korea Fish culture symposium (1)

NaGISA World Conference 2006 (1)

Asia-Pacific Genomics Symposium 2006 (2)

Autumn Meeting of the Japanese Society of Fisheries Science (1)

The 2007 Annual Meeting of the Nippon Nougai-Kagaku Gakkai (2)

The 2007 Annual Meeting of the Japanese Society of Fisheries Science (5)

### **A-3. Off-campus activities**

#### ***Research grants***

Toyohara, Haruhiko: Grant-in-Aid for Scientific Research (B)(2), Studies on fish muscle tenderization-analysis of transgenic red sea bream (Head: Toyohara); Grant-in-Aid for Scientific Research, Studies on biomineralization mechanism focusing on bivalve mantle (Head: Toyohara); The Bio-Design project from the ministry of Agriculture, Forestry and Fisheries, Japan; Research aid from Nippon Suisan Kaisha (Head: Toyohara), Salt Science Foundation, Cellulose decomposing ability of intertidal benthos(Head: Toyohara)

### **A-4. International cooperations and overseas activities**

Toyohara, Haruhiko: Japan-Thailand Symposium (invited speaker), JAMSTEC Research Program, Pan-Pacific Conference on Marine Biotechnology (Chairman)

## **B. Education activities (2006.4~2007.3)**

### **B-1. On-campus teaching**

a) Courses given

*Undergraduate level:* Basic Applied Biosciences II (Toyohara), Biotechnology-New Strategy in Agriculture- (Toyohara), Outline of Bioresource Science III (Toyohara), Molecular Biology (Toyohara), Practical Course in Marine Bioscience and Technology I (Toyohara), Cell Technology of Marine Organisms (Toyohara), Laboratory Course in Bioresource Science I and II (Toyohara), Introduction to Foreign Literature in Bioresource Science III (Toyohara), Seminar in Marine Bio-production (Toyohara), Marine Biology (Toyohara)

*Graduate level:* Seminar in Function of Marine Organisms (Toyohara), Laboratory Course in Function of Marine Organisms (Toyohara)

### **B-2. Off-campus teaching, etc.**

Toyohara, Haruhiko: Lecture (JAICA)

### **B-3.Overseas teaching**

Lecture in Prince of Songklar University

### **C. Other remarks**

#### ***Committees in the Faculty***

Toyohara, Haruhiko: Committee on Biological Education in Kyoto University. Committee on Genetically Engineered Fish, Committee of Evaluation of Post-Doctoral Fellows in JASP, Best paper award of Fisheries Science, Best poster award of Marine Biotechnology Conference