2.4.16 Laboratory: Marine Biological Function

Member: Associate Professor Toyohara, Haruhiko, Ph.D.

Assistant Professor Masati, Kinoshita, Ph.D.

Doctor's program 2

Master's Program 10

Undergraduate 4

Other 2

Researcher 1

A. Research Activities (2009.4-2010.3)

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, etylacetate, 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.
- 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 biominerarization process and isolate two genes that showed the homology with spider silk proteins.

d) Development of transgenic medaka as a vertebrate model

To evalulate the effecs of nano-particles on medaka (small freshwater teleost), we tested permiability of latex nano-particle. It was found that the particle reached brain throught blood-brain barrier. We also investigate the function of MATP gene, which is thought to concern in development of melanophore, with transgenic technology.

A-2. Publications and presentations

a) Publications

Books

- Medaka: Biology, Management, and Experimental Protocols. Edited M. Kinoshita, K. Murata, K. Naruse, and M. Tanaka. Wiley-Blackwell, Iowa, USA (2009)

Original Papers

- Fukamachi, S., M. Kinoshita, K. Aizawa, S. Oda, A. Meyer, H. Mitani: Dual control by a single gene of secondary sexual characters and mating preferences in medaka. BMC Biology 7; 64, 2009
- Sawatari, E., R.Seki, T. Adachi, H. Hashimoto, S. Uji, Y. Wakamatsu, T. Nakata, M. Kinoshita: Overexpression of the dominant-negative form of myostatin results in doubling of muscle-fiber number in transgenic medaka (Oryzias latipes). Comp. Biochem. Physiol. 155; 183-189, 2009
- Hano, T., Y. Oshima, M. Kinoshita, M. Tanaka, Y. Wakamatsu, K. Ozato, M. Nassef, Y. Shimasaki, T. Honjo: In ovo nanoinjection of nonylphenol affects embryonic development of a transgenic see-through medaka (Oryzias latipes), olvas-GFP/STII-YI strain. Chemosphere 77; 1594–1599, 2009
- Fukamachi, S., T. Yoda, A. Mayer, M. Kinoshita: Effects of constitutive expression of somatolactin alpha on skin pigmentation in medaka. Gene 442; 81-87, 2009
- Tonoyama, Y., D. Anzai, A. Ikeda, S. Kakuda, M. Kinoshita, T. Kawasaki, S. Oka: Essential role of beta1,4-galactosyltransferase 2 during medaka (Oryzias latipes) gastrulation. Mechanisms of Development 126; 580-594, 2009
- Murata, K., S. Degmetich, M. Kinoshita, E. Shimada: Expression of the congenital heart disease 5/tryptophan rich basic protein homologue gene during heart development in medaka fish, Oryzias latipes. Dev Growth Differ 51(2); 95-107, 2009

- Shimada, E.,M. Kinoshita, K. Murata: Expression of cardiac myosin light chain 2 (mcmlc2) during embryonic heart development in medaka fish, Oryzias latipes, and phylogenetic relationship with other myosin light chains. Develop. Growth Differ, 51; 1-16, 2009
- Kinoshita, M., G. Okamoto, T. Hirata, A. Shinomiya, T. Kobayashi, Y. Kubo, H. Hori, A. Kanamori: Transgenic medaka enables easy oocytes detection in live fish. Mol Reprod Dev 76; 202-207, 2009
- Suzuki, G., Hayashibara, T., Toyohara, H: Role of post-settlement mortality in the establishment of Acropora reef slope zonation inIshigaki Island, Japan. Galaxea 11; 13-20, 2009.
- Sakamoto, K and Toyohara, H: A comparative study of cellulose and hemicellulase activities of brackish water clam Corbicula japonica with those of other marine Veneoida bivalves. J Exp. Biol. 12; 2812-2818, 2009.
- Sakamoto, K and Toyohara, H: Putative endogenous xylanase from brackish-water clam Corbicula japonica. Comp. Biochem. Physiol B. 154; 85-92, 2009.
- Sakamoto, K and Toyohara, H: Molecular cloning of glycoside hydrolase family 45 cellulase genes from brackish water clam Corbicula japonica. Comp. Biochem. Physiol B. 152; 390-396, 2009.
- Sakamoto, K., Uji, S., Kurokawa, T. and Toyohara, H: Molecular cloning of endogenous β-glucosidase from common Japanese Brackish water clam Corbicula japonica. Gene, 435; 72-79, 2009.

Reviews

- Toyohara, H., Takahashi, J., Tohata K., Takagi M. and Kishida T: Spider silk in the shell, Earth, 31: 588-593, 2009.

b) Conference and seminar papers presented

- Japan-Korea benthic animal symposium (3)
- Autumn meeting of Japan Fisheries Science autumn (3)
- Meeting of Japan Fisheries Science (3)
- Meeting of Japan molecular biologists (1)
- Marine biotethnology conference (1)
- Annual meeting of Japanese Society of Developmental Biologysts (1)
- Annual meering of benthos (1)

- International Oryzias Meeting (1)
- JSPS Core University Program Meeting in Vietnam (1)
- World Ocean Conference, International Symposium on Ocean Sdience, Technology and Policy. (1)
- JSPS Core University Program Meeting in Thai (1)

A-3.Off-campus activities

Membership in academic societies

- Kinsohita Masato : The Japanese Society of Fisheries Science, The Molecular Biology Society of Japan, The Zoological Science of Japan
- Toyohara Haruhiko : The Japanese Society of Fisheries Science (Editor), The Japanese traditional food society (office)

Research grants

- 1. Grants-in-aid for Scientific Research(KAKENHI)
- Scientific Research (B): Kinoshita Masato: Detection of sex differntiation factor with female germ cell labbeled transgenic fish
- Challenging Exploratory Research : Kato Keitaro(Collaborator; Toyohara, Kinoshita) : Development of transgenic technique for marine fish
- Challenging Exploratory Research : Haruhiko Toyohara : Studies on the olfactory receptor of marine invertebrates
- Scientific Research (B): Haruhiko Toyohara: Biochemcal studies on the meiobenthos in the wet lands

A-4.International cooperation and overseas activities

International meetings(country,roles)

- Kinoshita, Masato Ph. D.: International symposium of Oryzias fish (Thailand, Organizer, Oral presenter), World Ocean Conference, International Symposium on Ocean Sdience, Technology and Policy (Indonasia, Oral Presenter)

B.Educational Activities(2009.4-2010.3)

B-1.On-campus teaching

a) Courses given

- Undergraduate level: Fundamentals for the laboratory course in bioresourse science

(Toyohara, Kinoshita), Practical course in marine bioscience and

technology II (Toyohara, Kinoshita), Laboratory course in

bioresource science I. II (Toyohara, Kinoshita), Seminor in marine

bio-production (Toyohara, Kinoshita), Graduation thesis

(Toyohara, Kinoshita), Biotechnology-New Strategy in Agriculture-(Toyohara), Cell Technology of Marine Organisms (Toyohara), Cell Biology II (Toyohara), Cell Biology I (Toyohara), Marine Biology

(Toyohara)

- Graduate level: Seminor in marine biological function (Toyohara, Kinoshita),

Laboratory course in marine biological function (Toyohara,

Kinoshita)

B-2.Off-campus teaching etc.

Part-time lecturer

- Kinoshita Masato: Kyoto Prefectural University of Medicin, Faculty of Medicine, Iwakuraminami primary school, Special Science Lecture

Open lectures, etc.

- Kinoshita Masato: Zeze High School, Special lecture in Kyoto University

B-3.Overseas teaching

<u>International students</u>

- International students: Research Students 2 (China)

Lectures and seminars

- Kinoshita Masato

Environmental Science(Lecturer): Thanomsak Boonphakdee, Brapha University(Thailand)
Application of ftransgenic medaka in environmental study(Lecturer): Inneke Rumengan,
Sam Ratulongi University(Indonesia)

Ecological function of the wet lands(Lecturer): Japan-Korea benthos conferences(Korea)