Chair Bioorganic and Biophysical Chemistry

2.3.10 Laboratory : Biofunction Chemistry

Member :	Professor	Hideto Miyoshi, Dr. Agric. Sci.
	Assistant Professor	Masato Abe, Dr. Agric. Sci.
	Assistant Professor	Masatoshi Murai, Dr, Agric. Sci.
	Master's Program	8
	Undergraduate	3

A. Research Activities (2010.4-2011.3)

A-1. Main Subjects

a) Bioorganic chemical study for elucidating mitochondrial complex I

Proton-translocating NADH-ubiquinone oxidoreductase (complex I) is the initial enzyme of the mitochondrial respiratory chain. It couples the transfer of two electrons from NADH to ubiquinone to the translocation of four protons across the inner mitochondrial membrane. The enzyme is composed of 45 different subunits with a total molecular mass of about 1 MDa. Because of complexity of the enzyme, our knowledge about the molecular structure and the catalytic mechanism is still highly limited. The aim of our research is to get insights into the structural and functional features of complex I through syntheses of various molecular probes and studies on their inhibition mechanism.

On the basis of structure-activity studies on complex I inhibitors, we have designed and synthesized radioisotope-tagged photoaffinity labeling probes for identification of the binding sites in complex I. We have revealed that the ND1 subunit, which is located in the membrane domain, constructs the inhibitor binding domain and plays an important role in the energy coupling in complex I.

b) Synthesis of functional ubiquinone probes and its biochemical application

Ubiquinone is an important electron-carrier in the respiratory chain of most eukaryotic and prokaryotic organisms. Recently, another roles of ubiquinone in the cell, such as antioxidant and redox regulation, have been suggested. In order to elucidate overall function of ubiquinone in the cell as well as its function in the respiratory enzymes, we are synthesizing various biotinylated ubiquinone probes aimed at biochemical experiments such as affinity purification, Western analysis and uptake assay

c) Construction of synthetic cardiolipin library

Cardiolipin, a phospholipid localized in the inner mitochondrial membrane, is believed to play important roles in the regulation of respiratory enzymes and the release of cytochrome c from mitochondria during the initial phase in apoptosis, while the details of the molecular mechanism is still unknown. We are constructing synthetic cardiolipin library, which can be used as powerful tools in various biochemical and biophysical experiments, to elucidate the biological functions in mitochondria.

A-2.Publications and presentations

a) Publications

Original Papers(including book-reviews)

- Kakutani, N., Murai, M., Sakiyama, N., and Miyoshi, H. (2010) Exploring the binding site of Δlac-acetogenin in bovine heart mitochondrial NADH-ubiquinone oxidoreductase, Biochemistry 49, 4794-4803

- Murai, M., Yamashita, T., Senoh, M., Mashimo, Y., Kataoka, M., Kosaka, H., Matsuno-Yagi, A., Yagi, T., and Miyoshi, H. (2010) Characterization of the ubiquinone binding site in alterative NADH-quinone oxidoreductase of Saccharomyces cerevisiae by photoaffinity labeling, Biochemistry 49, 2973-2980.

- Abe, M., Kitsuda, S., Ohyama, S., Koubori, S., Murai, M., and Miyoshi, H. (2010) Concise procedure for synthesis of cardiolipins having different fatty acid combinations, Tetrahedron Lett., 51, 2071-2073.

Reviews

- Murai, M. and Miyoshi, H.（2011）Kagaku 66, 72-73 (in Japanese).

b) Conference and seminar papers presented

- 35th Annual Meeting of Pesticide Society of Japan, 2 report

- 465th Kansai Branch Annual Meeting of Japan Society for Bioscience, Biotechnology and Agrochemistry; 1 report

- 36th Annual Meeting of Japan Bioenergetics Group; 3 reports

- 468th Kansai Branch Annual Meeting of Japan Society for Bioscience, Biotechnology and Agrochemistry; 1 report

- 36th Annual Meeting of Japan Pesticide Society of Japan; 3 report (Cancelled due to earthquake)

- Annual Meeting of Japan Society for Bioscience, Biotechnology and Agrochemistry; 4 report (Cancelled due to earthquake)

- 16th European Bioenergetics Conference (EBEC2010) ; 2 reports

A-3.Off-campus activities 1

Membership in academic societies

- H. Miyoshi, Dr. Agric, Sci. : Pesticide Science Society of Japan (councilor, editorial board member), Japan Society for Bioscience, Biotechnology, and Agrochemistry (editorial board member of english journal), Japan Bioenergetics Group (organizer)

A-3.Off-campus activities 2

Research grants

1. Grants-in-aid for Scientific Research(KAKENHI)

- Grant-in-Aid for Scientific Research (B) : H. Miyoshi : Exploring the function of membrane domain of mitochondrial complex-I using inhibitor probes.

- Grant-in-Aid for Young Scientists (B) : M. Abe : Functional study on cytochrome c-cardiolipin complex using synthetic cardiolipins.

- Grant-in-Aid for Young Scientists (Start-up) : M. Murai : Functional research on mitochondrial complex I based on the mode of action studies of the specific inhibitor.

- Grant-in-Aid for Exploratory Research : H. Miyoshi : Development of quinone-modified electrode for high-sensitive detection of respiratory enzymes

2.Other Research Grants

- The Uehara Memorial Foundation : M. Murai : Exploring the structure and function of mitochondrial NADH-ubiquinone oxidoreductase by photoaffinity labeling technique.

B.Educational Activities(2010.4-2011.3)

B-1.On-campus teaching

a) Courses given

- Undergraduate level : Introduction of applied life sciences IV (Miyoshi), Bioorganic chemistry II (Miyoshi), Laboratory course in bioorganic chemistry (Miyoshi, Abe, and Murai)

- Graduate level : Chemistry of biologically active compounds (Miyoshi), Biofunction chemistry seminar (Miyoshi), Experimental course of biofunctional chemistry (Miyoshi).

B-2.Off-campus teaching etc.

Part-time lecturer

- H. Miyoshi: Kinki University, Faculity of Agriculture (Special lecture on bio-organic chemistry)