

### 2.3.14 Laboratory : Plant Gene Expression

Member:	Professor	Yazaki, Kazufumi, Dr. Pharm. Sci.
	Associate Professor	Hayashi, Takahisa, Dr. Agric. Sci.
	Senior Lecturer	Kuroda, Hiroyuki, Dr. Agric. Sci.
	Doctor's program	2
	Master's Program	4
	Post-Doctoral fellow	3

#### **A. Research Activities (2009.4-2010.3)**

##### **A-1. Main Subjects**

###### a) outline

We are studying on the characterization of plant genes including woody plants which are involved in biosyntheses and transport of various valuable metabolites, e.g. secondary products, in plants, and also studying on the regulatory mechanism of the expression of those genes. The molecular breeding using those genes to establish novel woody plants, for instance phytoremediators to be applied for environmental biotechnology, is also our research targets. Individual research activity is as follows.

###### b) Molecular and cellular biology of secondary metabolism in higher plants and its application.

We are studying on the characterization of plant genes involved in biosyntheses of various secondary metabolites, e.g. isoprenoids and polyphenols, and elucidating the regulatory mechanism of the expression of those genes. In particular, we are interested in prenyltransferases accepting aromatic substrates leading to e.g. shikonin (red naphthoquinone) and coenzyme Q. Recent topic is the first identification of flavonoid-specific prenyltransferase in plants. As applied scientific approaches we are utilizing those genes to alter plant functions, e.g. molecular breeding of thermotolerant plants by isoprene synthase gene and alteration of fragrance of tree species by monoterpene synthases.

###### c) Molecular biology of transporter proteins in plants

Plants possess ca. 130 members of (ATP-binding cassette) ABC proteins. Some of them are reported to function as molecular pump for xenobiotics. In particular we are characterizing ABCB- and ABCG-subfamily members, i.e. their roles in the auxin transport in Arabidopsis,

and their involvement in the nodule formation in legume plants. Recent topic is the identification of nicotine transporter functioning at the vacuolar membrane in tobacco, which is a MATE-type transporter. As applied sciences using such transporter genes, we attempt to establish phytoremediation method to clean environment with transgenic plants

d) Cell wall and cellulose biosynthesis

Cell wall loosening: This study focuses on the structure and function of endo-1,4- $\beta$ -glucanase. Biosynthesis of cellulose in higher plants and in *Acetobacter xylinum*:: Molecular and cell biology of cellulose biosynthesis in higher plants and *Acetobacter xylinum*.

e) Molecular biology of intrinsic cDNA clones from woody plants

We are focusing on the cDNAs involved in pathogen-resistant traits, some of which are related to secondary metabolism and water stress in woody plants. Their translates and transcripts are respectively studying for the molecular machines and for making a diagnosis of the forest biosphere possible

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

a) Publications

Books

- Plant ATP-binding cassette proteins. In: Plant Nutrition 2nd edition. (edited by Matoh, Fujiwara, Ma), Bunei Publishing Co. (Tokyo) (in Japanese)

Original Papers

- Ohara, K., Matsunaga, E., Nanto, K., Yamamoto, K., Sasaki, K., Ebinuma, H., Yazaki, K., Monoterpene engineering in a woody plant *Eucalyptus camaldulensis* using a limonene synthase cDNA. *Plant Biotech. J.*, 8 (1), 28-37 (2010).

- Pomahacová B, Dusek J, Dusková J, Yazaki K, Roytrakul S, Verpoorte R., Improved accumulation of ajmalicine and tetrahydroalstonine in *Catharanthus* cells expressing an ABC transporter, *J. Plant Physiol.*, 166(13), 1405-1412 (2009).

- Ohara, K., Muroya, A., Fukushima, N., Yazaki, K., Functional characterization of LePGT1, a membrane-bound prenyltransferase involved in the geranylation of para-hydroxybenzoic acid. *Biochem. J.*, 421 (2), 231-241 (2009).

- Shitan, N., Morita, M., Yazaki, K., Identification of a nicotine transporter in leaf vacuoles of *Nicotiana tabacum*. *Plant Signaling & Behavior*, 4 (6), 530-532 (2009).

- Sasaki, K., Tsurumaru, Y., Yazaki, K., Prenylation of flavonoids by the biotransformation of yeast expressing plant membrane-bound prenyltransferase SfN8DT-1. *Biosci. Biotech. Biochem.*, 73 (3), 759-761 (2009).

- Kaida, R., Kaku, T., Baba, K., Hartati, S., Sudarmonowati, E. and Hayashi, T., Enhancement of saccharification by overexpression of poplar cellulase in sengon., J. Wood Sci. 55 435-440 (2009)
- Kaida, R., Kaku, T., Baba, K., Oyadomari, M., Watanabe, T., Hartati, S., Sudarmonowati, E. and Hayashi, T., Enzymatic saccharification and ethanol production of *Acacia mangium* and *Paraserianthes falcataria* wood, and *Elaeis guineensis* trunk., J. Wood Sci. 55 381-386 (2009).
- Kaku, T., Serada, S., Baba, K., Tanaka, F. and Hayashi, T., Proteomic analysis of the G-layer in poplar tension wood., J. Wood Sci. 55 250-257 (2009)
- Baba, K., Park, Y.W., Kaku, T., Kaida, R., Takeuchi, M., Yoshida, M., Hosoo, Y., Ojio, Y., Okuyama, T., Taniguchi, T., Ohmiya, Y., Teiji, Kondo, T., Shani, Z., Shoseyov, O., Awano, T., Serada, S., Norioka N., Norioka, S. and Hayashi T., Xyloglucan for generating tensile stress to bend tree stem., Mol. Plant 2 893-903 (2009).
- Kaida, R., Kaku, T., Baba, K., Oyadomari, M., Watanabe, T., Nishida, K., Kanaya, T., Shani, Z., Shoseyov, O. and Hayashi, T., Loosening xyloglucan accelerates the enzymatic degradation of cellulose in wood., Mol. Plant 2 904-909 (2009).
- Kaida R., Satoh Y., Bulone V., Yamada Y., Kaku T., Hayashi T., Kaneko T. , Activation of b-glucan synthases by wall-bound purple acid phosphatase in tobacco cells. , Plant Physiol.150:1822-30. 2009

#### Reviews

- Yazaki, K., Sasaki, K., Tsurumaru, Y., Prenylation of aromatic compounds, a key diversification of plant secondary metabolites. Phytochemistry, 70 (15-16), 1739-1745 (2009).
- Yazaki, K., Shitan, N., Sugiyama, A., Takanashi, K., Cell and molecular biology of ATP-binding cassette proteins in plants. Intl. Rev. Cell Mol. Biol., 276, 263-299 (2009).
- Kumazawa, S., Nakamura, J., Ohta, T., Yazaki, K., Miyagi, T., Fukumoto, S., Plant origin of Okinawan propolis. Kagaku-to-Seibutsu, 48 (1), 35-42 (2010). (in Japanese)
- Sugiyama, A., Shitan, N., Kuzuyama, T., Yazaki, K., Development of transgenic plants producing prenylated polyphenols. Bio Industry, 26 (1), 41-48 (2009). (in Japanese)

#### Reports

- Itoh, S., Yazaki, K., Alkaloid transporter protein functioning at vacuoles. BRAIN Techno News, vol. 135, 11-15 (2009). (in Japanese).

- Yazaki, K., Transporter involved in the accumulation of nicotine in tobacco leaves. Kagaku-to-Seibutsu, Vol., 135, 11-15 (2009). (in Japanese).
- Kuroda, H. , Matsuda,T., Hattori, M., Kuroda, K. , Molecular evaluation for the forest health in Japanese red pine. News letter of ISS, Vol.7, 7, 2009.

#### Patents

- Kyoto University, Api Co. Ltd., Yazaki, K., Prenyltransferase. Pat. No. 2010-22323、2010.2.4

#### b) Conference and seminar papers presented

- Plant Biology 2009: 1 report
- Terpnet 2009: 2 reports
- The 5th Takeda Science Foundation Symposium on PharmaSciences: 2 reports
- XIV International Congress on Molecular Plant-Microbe Interactions: 1 report
- Colloquium of the Priority Programme –Evolution of Metabolic Diversity–: 1 report
- Annual Meeting of Japanese Society for Bioscience, Biotechnology and Agrochemistry 2010 : 2 reports
- 51th Annual Meeting of Japanese Society for Plant Physiologists : 6 reports
- The 26th Annual Meeting of Japanese Society for Plant Cell and Molecular Biology : 3 reports
- 130th Annual Meeting of Pharmaceutical Sciences : 1 report
- 4th Meeting on Transporters : 2 report
- 19th Plant-Microbe Interaction : 1 report
- 27th Biotechnology Symposium : 1 report
- 51th Symposium on Natural Product Chemistry : 1 report
- 2nd The Hakata Symposium : 1 report
- 46th Plant Chemistry Symposium : 1 report
- 60th Annual Meeting of Japanese Wood Research Society: 1 report

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

##### Membership in academic societies

- Yazaki, Kazufumi, Dr. Pharm. Sci. : Japanese Society for Plant Physiologist (Secretary, Editor), The Japanese Society for Plant Cell and Molecular Biology (Secretary, Editor), Japan

Society for Bioscience, Biotechnology, and Agrochemistry (Board member), The Japanese Bioindustry Association (Editorial Board), Association of Bio Quinone (Executive Board)

#### Research grants

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

- Scientific Research on Priority Areas : Yazaki, Kazufumi : Transport of signal molecules by ABC proteins involved in the generation of lateral organs and the meristem regulation
- Scientific Research on Priority Areas : Yazaki, Kazufumi : LjPDR1, an ABC transporter involved in symbiotic nitrogen fixation and the regulation of nodulation
- Basic Research (B) : Yazaki, Kazufumi : Functional dissection and enzymatic engineering of flavonoid prenyltransferase family
- Basic Research (B) : Hayashi, Takahisa : Field trial of transgenic poplar for commercial utilization.
- Basic Research (B) : Hayashi, Takahisa : Transgenic trees: Overseas research of field trial.
- Challenging Exploratory Research : Kuroda, Hiroyuki : Research and development on the device for biopolymer extractions

##### 2. Other Research Grants

- METI Project: Yazaki, Kazufumi: Plant metabolic engineering with prenyltransferase genes
- Research Grant for Sustainability Science: Aoyama, Takashi (Yazaki, Kazufumi): Morphology and Function of Root Hairs Supporting Phosphate Absorption from Soil
- Mission Grant for Sustainable Humansphere: Arimura, Gen-ichiro (Yazaki, Kazufumi): Basic studies on transgenic plants expressing terpene synthase and prevention of herbivores.
- Mission Grant for Sustainable Humansphere: Sugimoto, Kohichi (Yazaki, Kazufumi): Chemical sensing by plants -molecular ecological study on the preception of volatile compounds-
- Mission Grant for Sustainable Humansphere: Ogushi, Takayuki (Yazaki, Kazufumi): Genetic diversity of woody species and their effects on the ecology system of arthropods.
- Research Grant for Sustainability Science: Hayashi, Takashi: Utilization assessment of biomass in Biwa Lake.
- Research Grant for Sustainability Science: Kuroda, Hiroyuki: Molecular evaluation for the forest health in Japanese red pine.

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

##### International meetings(country,roles)

- Yazaki, Kazufumi: Yazaki, K., Colloquium of the Priority Programme –Evolution of Metabolic Diversity– (invited speaker), Terpnet 2009 (organizer)

**B.Educational Activities(2009.4-2010.3)****B-1.On-campus teaching**

## a) Courses given

- Undergraduate level: Science of Sustainable Humanosphere (Shiotani, Tsuda, Yazaki), SI lecture, Diagnostics and Control of Humanosphere (Shiotani, Hashiguchi, Horinouchi, Yazaki, Honda, Umezawa, Sugiyama) , Plant Biochemistry II (Yazaki, Umezawa)
- Graduate level: Laboratory Course in Plant Gene Expression (Yazaki, Hayashi, Kuroda), Seminar in Plant Gene Expression (Yazaki, Hayashi, Kuroda), Plant Gene Expression (Yazaki)

**C.Other Remarks**

- Yazaki, Kazufumi, Dr. Pharm. Sci. : METI Plant Project Committee (Board member)