2.5.6 Laboratory: Environmental Mycoscience

Member:	Professor	Futai, Kazuyoshi
	Associate Professor	Tanaka, Chihiro
	Assistant Professor	Takeuchi, Yuko
	KU Visiting Professor	Wang, Jianguo
	Doctor's program	7
	Master's Program	8
	Undergraduate	2
	Other	2

A. Research Activities (2009.4-2010.3)

A-1. Main Subjects

a) Ecological studies on interrelationships among organisms involved in two forest epidemics 1. To elucidate infection mechanisms of pine wilt disease, several pure lines of pathogenic nemaotodes were established. Hence we will become able to analyze their pathogenicity more precisely. Pathogenic factors have been studied by proteomic analysis, and some pathogic-nematode specific proteins were discovered. While, host genes expressed after nematode infection have also been studied intensively. As for the pine wilt disease, field survey has been conducted on its way to reveal the role of the latent carrier trees in expanding this epidemics.

2. Another forest epidemic, Japanese Oak Wilt is also vectored by a tiny beetle, Platypus quercivora. To develop effective control methods, their reproductive system in host tree was investigated from the behavioral viewpoints, and their subsociality shown in sharing social roles among male, female, and larvae was elucidated. Also their food source, yeast species, were studied from community ecological viewpoint, and several prevalent species grown on the wall of tunnel systems, and/or found in vector's mycangia were recorded. Various microorganisms isolated from dead oak trees and beetles obtained from such trees were examined their ability to kill the beetles, and the application methodes and their applicability of such microorganisms to control the disease have been examined.

b) Biochemical and ecological genetics on fungi

1. The two classes of fungicides, Dicarboximides and Phenylpyrroles are effective and used for control of several phytopathogenic fungi. The modes of the actions of these fungicides were unclear. However, our studies using the mutants of Cochliobolus heterostrophus revealed that these fungicides improperly activate the filamentous-fungus specific high-osmolarity stress signaling pathway. This signaling pathway has a great attention as a target of the fungicides. The pathway is involved not only in the adaption for high osmotic environment but also in the adaption for the host plant. The pathway seems to have a critical role in the lifecycles of plant pathogenic filamentous fungi. In this year, we attempted to study crosstalk of the high-osmolarity stress signaling pathway and other MAPK signaling pathways. We developed Stel1 disruptant, which encodes MAPKK in the Chk1-MAPK pathway. Δ Chstel1 strains showed defects in conidiation, sexual development, melanization and the formation of appressoria. These mutants were significantly less virulent on corn plants than the wild type. Δ Chstell strain showed severe sensitivity to oxidative stress, hydrogen peroxide, and heavy metals, cupric or ferric cations. Δ Bmhog1 strains, mutants of the HOG1-type MAPK, did not show sensitivity to these forms of stress. The results suggested the two MAPK signaling pathway were independently involved in stress responces in this fungi.

2. In eukaryotes, copper-transporting ATPases deliver copper to various copper-containing proteins in the trans-golgi network. This study identified a copper transporting ATPase gene BcCcc2 in a fungus pathogenic to plants, Botrytis cinerea. We investigated the biological roles of BcCCC2 by generating null mutants for BcCcc2. Melanization, conidiation and the formation of sclerotia were severely affected in \triangle BcCcc2 mutants. Moreover, a pathogenicity assay using tomato leaves and carnation petals revealed the mutants to be nonpathogenic. Further analysis indicated that they formed fewer appressoria and infection cushions than the wild type. These structures were aberrant in morphology and in many cases, had a significantly reduced ability to penetrate the plant epidermis. An assay also indicated that \triangle BcCcc2 mutants were defective in infection through wounds. BcCCC2 is necessary not only for penetrating a host but also for fungal growth within plant tissues. Our results also imply that B. cinerea requires copper-containing proteins for infection that are inactive in the absence of the copper-transporting ATPase BcCCC2.

c) Studies on symbiotic relationships between microorganisms and trees

Many tree species in temperate zone have mycorrhizal relationships with fungi. The effects of mycorrhizal fungi on host trees have been studied from two points of view; (1) mitigation of water stress, and (2) improvement of host resistance. The roles of endophytic actinomycetes have also been investigated and their effects on seedling establishment and on its growth were elucidated.

A-2.Publications and presentations

a) Publications

Original Papers

 Izumitsu, K., A. Yoshimi, A. Hamada, A. Morita, Y. Saitoh and C. Tanaka:
 Dic2 and Dic3 loci confer osmotic adaptation and fungicidal sensitivity independent of the HOG pathway in Cochliobolus heterostrophus. Mycological Research 113; 1208-1215, 2009

- Izumitsu, K., A. Yoshimi, D. Kubo, A. Morita, Y. Saitoh and C. Tanaka, C:

The MAPKK kinase ChStel1 regulates sexual/asexual development, melanization,

pathogenicity, and adaptation to oxidative stress in Cochliobolus heterostrophus. Current Genetics 55; 439-448, 2009

- Kataoka, R. and K. Futai:

A new mycorrhizal helper bacterium, Ralstonia species, in the ectomycorrhizal symbiosis between Pinus thunbergii and Suillus granulatus. Biol. Fertil. Soils 45; 315-320, 2009

- Kataoka, R., Z.A. Siddiqui, T. Taniguchi and K. Futai:

Quantification of Wautersia [Ralstonia] basilensis in the mycorrhizosphere of Pinus thunbergii Parl. and its effect on mycorrhizal formation. Soil Biol. Biochem. 41; 2147-2152, 2009

- Kataoka, R., T. Taniguchi and K. Futai:

Fungal selectivity of two mycorrhiza helper bacteria on five mycorrhizal fungi associated with Pinus thunbergii. World J. Microbiol. Biotechnol. 25; 1815-1819, 2009
Muroi, A., A. Ishihara, C. Tanaka, A. Ishizuka, J. Takabayashi, H. Miyoshi and T. Nishioka:

Accumulation of hydroxycinnamic acid amides induced by pathogen infection and identification of agmatine coumaroyltransferase in Arabidopsis thaliana. Planta 230; 517–527, 2009

- Saitoh, Y., K. Izumitsu and C. Tanaka:

Phylogenetic analysis of heavy-metal ATPase in fungi and characterization of the copper-transporting ATPase of Cochliobolus heterostrophus. Mycological Research 113; 737-745, 2009

- Shinya, R., Y. Takeuchi, N. Miura, K. Kuroda, M. Ueda and K. Futai: Surface coat proteins of the pine wood nematode, Bursaphelenchus xylophilus: profiles of stage and isolate specific characters. Nematology 11; 429-438, 2009 - Siddiqui, Z.A. and K. Futai:

Biocontrol of Meloidogyne incognita on tomato using antagonistic fungi, plant-growth-promoting rhizobacteria and cattle manure. Pest Management Science 65; 943-948, 2009

- Takeuchi, Y. and K. Futai:

Diagnosis and quantification of the pine wood nematode, Bursaphelenchus xylophilus (Steiner and Buhrer), in wood of Pinus thunbergii with real-time PCR. Nematol. Res. 39; 9-16, 2009

- Taniguchi, T., R. Kataoka, S. Tamai, N. Yamanaka and K. Futai:

Distribution of bacterial species in soil with a vegetational change from Japanese black pine (Pinus thunbergii) to black locust (Robinia pseudoacacia). Microbes Environ. 24; 246-252, 2009

- Taniguchi, T., R. Kataoka, S. Tamai, N. Yamanaka and K. Futai:

Distribution of ectomycorrhizal and pathogenic fungi in soil along a vegetational change from Japanese black pine (Pinus thunbergii) to black locust (Robinia pseudoacacia). Mycorrhiza 19; 231-238, 2009

Reviews

Tanaka, C., K. Izumitsu, K. Yoshimi, K. Shimizu, M. Kimura and T. Motoyama:
Osmotic signaling pathway of plant pathogenic fungi: Kagaku to Seibutsu 47; 644–650, 2009 (in Japanese)

b) Conference and seminar papers presented

- IUFRO 2009 International Symposium on Pine Wilt Disease: 2 papers
- The 53rd Annual Meeting of the Mycological Society of Japan: 1 paper
- The 17th Annual Meeting of the Japanese Nematological Society: 3 papers
- The 54th Annual Meeting of the Japanese Society of Applied Entomology & Zoology:
- 2 papers
- The 120th Annual Meeting of the Japanese Forestry Society: 4 papers

- The Annual Meeting of the Fungal Molecular Biology Society of Japan, 2009: 2 papers

- The Annual Meeting of the Phytopathological Society of Japan, 2009: 1 paper

A-3.Off-campus activities

Membership in academic societies

- Futai, Kazuyoshi, D.Agric.Sci : The Japanese Nematological Society (councilor), Tree Health Research Society (member of editorial board)

- Tanaka, Chihiro, D. Agric. Sci. : The Mycological Society of Japan (councilor, member of database committee), Phytopathological Society of Japan (member of editorial board)

- Takeuchi, Yuko, D. Agric. Sci. : The Japanese Nematological Society (councilor)

Research grants

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

Scientific Research (B2) : Osawa, Naoya (Collaborator; Tanaka, Chihiro) : Applied ecological syudy on management system for Japan-originated invadors in foreign contries
Young Scientists (B) : Takeuchi, Yuko : Molecular biological study of the pathogenic nematode-host plant interactions of pine wilt disease

2. Other Research Grants

- Grant in Aid for Agricultural Research and Technology from Agriculture, Forestry and Fisheries Research Council: Ohta, Akira (Collaborator; Tanaka, Chihiro): Development of cultivation methods for mushroom production of Matsutake allies.

A-4.International cooperation and overseas activities

Membership in academic societies

- Futai, Kazuyoshi, D.Agric.Sci: European Society of Nematologists (representative of Japanese members), IUFRO (a deputy of the session of Pine Wilt Disease)

International meetings(country,roles)

- Futai, Kazuyoshi, D.Agric.Sci: International Symposium on Pine Wilt Disease (a member of Scientific committee, a cordinator)

International joint research, overseas research surveys

- A study on fungal flora in a tropical and monsoon South-East Asia; Tanaka, Chihiro; Malaysia

- Studies on exotic ectomycorrhizal fungi invaded into New Zealand native forest; Tanaka, Chihiro; New Zealand

Visiting Research Scholars

- KU Guest Professor 1 (China)

B.Educational Activities(2009.4-2010.3)

B-1.On-campus teaching

a) Courses given

- Undergraduate level:	Microbes in Ecosystem (Futai), Seminar in Environmental	
	Microbiology (Futai, Tanaka), Laboratory course in Biological and	
	Environmental Science III, IV (Futai, Tanaka, Takeuchi), Outline of	
	Bioresource Science IV (Futai, Tanaka), Microbiology (Futai,	
	Tanaka), Pesticide Science (Tanaka).	
- Graduate level:	Seminar in Environmental Mycoscience (Futai, Tanaka, Takeuchi),	
	Research in Environmental Mycoscience (Futai, Tanaka, Takeuchi),	

B-3.Overseas teaching

International students

- International students : Doctral 2 (China 1, Indonesia 1)