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Targeted approaches in metabolic science of forest plants and microorganisms

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It is becoming more important to establish a sustainable society, which depends on renewable resources. Because wood biomass is the most abundant renewable resource, studies of wood formation is the key to improve forest biomass production. In this context, we are involved in analyzing metabolic functions of forest plants and microorganisms from a wide variety of aspects, including organic chemistry, biochemistry, molecular biology, and metabolomics, in order to conduct basic investigations contributing cultivation and protection of forest resources.

1. Molecular cloning of novel genes encoding enzymes of phenylpropanoid biosynthesis

In the post-genomic era, to understand mechanisms for metabolisms in forest plants and microorganisms, the integration of comprehensive or holistic analyses such as transcriptomics, proteomics and metabolomics and targeted analyses are critically important.

The cinnamate/monolignol pathway supplies precursors for various phenylpropanoid compounds such as lignins, lignans, and norlignans, which are involved in secondary xylem formation composed of cell-wall and heartwood formation of herbaceous and woody plants. The lignan pathway which follows the cinnamate/monolignol pathway is involved in heartwood formation and production of bioactive compounds. Thus, these pathways play central roles in plant secondary metabolism.

We have been working on comprehensive or network analysis of gene expression involved in secondary xylem formation. In addition, from the aspects of the targeted approach, we have cloned two novel genes. One is a novel *O*-methyltransferase (OMT) catalyzing methylation of a lignan, matairesinol, giving rise to arctigenin. This is the first example of molecular cloning of a cDNA encoding a lignan OMT. In addition, we have cloned cDNAs encoding a norlignan synthase, which catalyzes the formation of hinokiresinol from *p*-coumaryl *p*-coumarate. Both genes or their homologs are involved in heartwood formation, and these enzymes are can be used as clues to help us elucidate heartwood formation mechanisms.

2. Mechanisms for organic acid metabolism of wood-rotting fungi and ectomycorrhizal fungi

Biodegradation of wood components by wood-rotting (WR) fungi including white- and brown-rot basidiomycetes is important as a first process leading to humus production, which in turn contributes greatly to sustainable forest ecosystems. Oxalate excreted from WR fungi plays a wide variety of roles in the degradation owing to its various chemical natures. We have proposed that oxalate metabolism is an important biochemical device to produce energy for fungal growth of WR fungi. Firstly, we reported that oxalate decarboxylase and formate dehydrogenase play an important role in decomposing oxalate to produce NADH as an energy source during vegetative growth of white-rot fungus *Cereporiopsis subvermispora*. Secondly, we proposed that peroxisomal glyoxylate (GLOX) cycle cooperatively play a role with mitochondrial TCA cycle in oxalate fermentation by which brown-rot fungus *Fomitopsis palustris* acquires energy for fungal growth on glucose. To the best of our knowledge, this is a first example to show peroxisomal GLOX cycle in glucose-grown mycelia. Furthermore, it was postulated that in glucose-grown *F. palustris* some of the common metabolites of the peroxisomal GLOX and mitochondrial TCA cycles are transported between peroxisomes and mitochondria in a constitutive and cooperative manner in terms of the oxalate biosynthesis.

Cytochemical and molecular approaches for enzymes and transporters involved in organic acid metabolism are being investigated for WR fungi. Furthermore, comprehensive study for elucidation of regulatory mechanisms for organic acid metabolism in WR and ectomycorrhizal fungi has just begun.