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 RECENT RESEARCH ACTIVITIES

Structure, biosynthesis, and bioengineering of lignocellulose and phenylpropanoid metabolites for future biorefinery

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It is becoming increasingly important to establish a sustainable society by reducing our excessive reliance on fossil resources and mitigate global climate change. As lignocellulosic biomass represents the most abundant renewable and carbon-neutral resources on earth, technologies to improve their utilizations are key for realizing the goal. In this context, we investigate the structure, biosynthesis and bioengineering of lignocellulose using various model plants and biomass crops. In addition, we are interested in understanding the biosynthesis of plant-derived phenylpropanoid metabolites with useful biological activities. Our program typically integrates research ideas and approaches based on chemistry, biochemistry, and plant molecular biology.

Among a wide variety of biomass feedstocks, grass biomass crops, such as *Erianthus*, *Sorghum*, sugarcane and bamboo, have attracted particular attention due to their high biomass productivity and superior environmental adaptability. To explore new breeding strategies to improve the production of useful fuels and materials from grass biomass, we seek to develop transgenic rice plants that produce biomass with improved utilization properties. Our research particularly focuses on manipulating lignin, a phenylpropanoid polymer accounting for 15-30 wt% of lignocellulosic biomass. We have developed various rice transgenic lines in which specific genes encoding enzymes and transcription factors functioning in the lignin biosynthetic pathway are down- and/or up-regulated. Some of our developed transgenic lines display notably improved biomass utilization properties. In parallel, we are also working on selective breeding of grass crop varieties, such as *Erianthus* spp. and *Sorghum* spp., with superior lignins suited for bioenergy and biomaterial productions. In addition, aiming at biological production of useful phytochemicals, we have been characterizing plant and microbial enzymes/genes involved in formations of bioactive phenylpropanoids such as lignans and norlignans. Our recent projects include elucidation of the biosynthesis of antitumor podophyllotoxin in *Anthriscus sylvestris*, unravelling crystal structures of hinokiresinol synthases, unique enzymes responsible for the enantioselective formation of bioactive norlignans, and identification of enzymes/genes involved in the formation of estrogenic mammalian lignans (enterolignans) via human intestinal bacteria.

Selected Publications (FY2018)

- [1] Takeda Y et al. (2018) Downregulation of p-COUMAROYL ESTER 3-HYDROXYLASE in rice leads to altered cell wall structures and improves biomass saccharification. *The Plant Journal* 95: 796-811.
- [2] Miyamoto et al. (2018) Comparative analysis of lignin chemical structures of sugarcane bagasse pretreated by alkaline, hydrothermal, and dilute sulfuric acid methods. *Industrial Crops and Products* 121: 124-131.
- [3] Umezawa T. Lignin modification *in planta* for valorization. *Phytochemistry Reviews* 17: 1305-1327.
- [4] Takeda Y et al. (2019) Lignin characterization of rice CONIFERALDEHYDE 5-HYDROXYLASE loss-of-function mutants generated with the CRISPR/Cas9 system. *The Plant Journal* 97: 543-554.
- [5] Takeda Y et al. (2019) Comparative evaluations of lignocellulose reactivity and usability in transgenic rice plants with altered lignin composition. *Journal of Wood Science* 65:6.
- [6] Tobimatsu Y. and Schuetz M. (2019) Lignin polymerization: how do plants manage the chemistry so well? *Current Opinion in Biotechnology* 56:75-81.
- [7] Miyamoto T et al. (2019) OsMYB108 loss-of-function enriches p-coumaroylated and triclin lignin units in rice cell walls. *The Plant Journal*, in press (DOI: 10.1111/tpj.14290).
- [8] Lam PY et al. (2019) Recruitment of specific flavonoid B-ring hydroxylases for two independent biosynthesis pathways of flavone-derived metabolites in grasses. *New Phytologist*, in press (DOI:10.1111/nph.15795).
- [9] Suzuki S et al. (2019) *De novo* transcriptome analysis of needles of *Thujopsis dolabrata* var. *hondae*. *Plant Biotechnology*, in press.