***Introduction of chemically labile units into lignin***

Lignin is a complex polymer found in plant secondary walls that plays an important role in mechanical support, water transport, and stress responses. In many applications, lignin has to be removed in order to access the cellulose and other polysaccharides for conversion into bioproducts or biofuels. Bacteria-derived enzymes that can modify specific lignin substrates are potential targets to engineer plants for better biomass processability. GLBRC Researchers and collaborators in twelve different organizations across four countries investigated whether a specific bacterial enzyme known to degrade lignin could be expressed *in planta*. *Arabidopsis thaliana*, a model plant, was engineered to express Cα-dehydrogenase (LigD) from *Sphingobium* sp. SKY-6, one of the best-characterized lignin degrading enzymes. This enzyme has been shown, *in vitro*, to oxidize the most abundant linkage in native lignin (β-O-4) to a more labile (α-keto) analogue. Enzyme activity in transgenic plants was determined using RT-PCR and Western blot analysis; UPLC-MS was used for phenolic profiling; 2D NMR was used to determine cell wall composition and lignin structure. The transformation was successful and researchers were able to show that the lignin structure was modified in the desired manner, albeit at a fairly low level. LigD was successfully expressed and enzyme activity detected in extracts of transgenic species. LigD was shown to oxidize a wider range of β-O-4-linked compounds than previously known suggesting that the enzyme tolerates variations in side-chains of native lignin transgenic lines, something that was not known before this study. It was also demonstrated that although the total lignin in transgenic lines was similar to that in WT, the transgenic plants had more of the desired α-keto linkages. And, although there was no detectable change in saccharification efficiency between the transgenic lines and the wild-type control, the ability to express a bacterial enzyme *in planta*, and have that enzyme act upon the lignin polymer, is a significant step towards generating dedicated bioenergy crops.

**Reference:
Tsuji Y., Vanholme R., Tobimatsu Y., Ishikawa Y., Foster C. E., Kamimura N., Hishiyama S., Hashimoto S., Shino A., Hara H., Sato-Izawa K., Oyarce P., Goeminne G., Morreel K., Kikuchi J., Takano T., Fukuda M., Katayama Y., Boerjan W., Ralph J., Masai E. and Kajita S.** (2015) Introduction of chemically labile substructures into Arabidopsis lignin through the use of LigD, the Cα-dehydrogenase from *Sphingobium* sp. strain SYK-6. **Plant Biotechnol. J**., doi*:* [*10.1111/pbi.12316*](http://dx.doi.org/10.1111/pbi.12316)

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