

## Towards New Degradable Lignin Types

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### Abstract

Lignin is an aromatic heteropolymer abundantly present in secondarily-thickened plant cell walls. It is a major limiting factor in the conversion of lignocellulosic biomass to liquid biofuels. In dicots, the lignin polymer is built up by the combinatorial radical coupling of mainly coniferyl and sinapyl alcohol, although a range of minor units are also present in the polymer. Radical coupling results in a variety of chemical bonds, the frequency of which depends on the relative abundance of the various monomers, on the chemical characteristics of the monomers, and the local environment in the cell wall [1-5]. For end use applications, such as the conversion of lignocellulosic biomass to fermentable sugars in the process to bioethanol, cell walls would ideally contain less lignin, and lignin rich in bonds that are easily cleaved. The goal of the project is to identify natural products (called “target molecules”) that can be biosynthesized in energy crops, translocated through the plasma membrane and cross-coupled with lignin units such that the final lignin polymer is more susceptible to chemical cleavage, or is more hydrophilic, or is less cross-linked with hemicelluloses. Ideally, the structures of the target molecules are very similar to traditional monolignols so that they can be exported to the wall using the same transport system. The project is divided into four tasks. Task 1 aims at defining which molecules are good targets for engineering and targeting to plant cell walls. We have compiled a list of such target molecules. Task 2 aims at cloning biosynthetic genes for target molecules and overexpressing these in bioenergy crops. We have identified an Arabidopsis gene that, upon down-regulation, steers the flux through the phenylpropanoid pathway into the biosynthesis of ferulate-derivatives. This is interesting, because if we can ship the ferulate to the wall, it may give rise to labile acetal bonds. Task 3 is more challenging, and aims at rerouting a selected number of target molecules, of which the biosynthetic pathway and subcellular localization are already partially known, to the cell wall in transgenic plants. Because several of the proposed target molecules are located in the vacuole, we have identified a putative transporter that is involved in translocation/retention of several target molecules into the vacuole. Transgenic plants silencing this transporter have been generated and await analyses. In addition, we have now a collection of Arabidopsis knock-out mutants in putative transporters genes, the latter being differentially expressed in a collection of lignin mutants, thus making them excellent candidates for a role in transport of phenylpropanoid derivatives into the vacuole. We also have demonstrated the vacuolar location of some of the target molecules. Task 4 is the most risky, and

aims at identifying biosynthetic pathways for promising target molecules by a combination of genetics and metabolomics. To this end, we have metabolically profiled Arabidopsis ecotypes and identified several of our target molecules in this model system, opening perspectives to clone their biosynthetic pathways using genetic strategies.

## References

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