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Identification of Genes Involved in Acetylation of Cell Wall Polysaccharides in *Arabidopsis thaliana*

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Acetylation of cell wall polysaccharides has long been observed in various plant species; however, the enzymes involved in the acetylation have thus far not been identified. Both pectins and hemicelluloses are acetylated to various degrees. While the *in vivo* role of polysaccharide acetylation is still unclear, it is known to affect biofuel yield from lignocellulosic biomass due to inhibition of enzymatic degradation of the polysaccharides and fermentation by microorganisms. Therefore, a decreased level of acetate esters in lignocellulosic biomass may increase the efficiency of biofuel production. JBEI is therefore investigating the mechanism of polysaccharide acetylation and assessing the possibility of modifying acetylation level *in planta*.

We have analyzed four *Arabidopsis* homologues of a protein known to be involved in polysaccharide acetylation in a fungus. *Arabidopsis* mutants with insertional mutagenesis in the respective genes were identified, and we found that at least one of the mutants, designated *reduced wall acetylation* (*rwa1*, *rwa2*, *rwa3* and *rwa4*) had decreased levels of acetylated cell wall polymers. Two independent alleles of *rwa2* mutants were examined by analyzing alcohol insoluble residues extracted from leaves. Extracts treated with 0.1M NaOH released about 20% lower amounts of acetic acid when compared to wildtype. Interestingly, the monosaccharide composition of the cell wall polysaccharides in *rwa2* was not altered. Current efforts are aimed at determining which acetylated polysaccharides are affected in the *rwa2* mutants. There was no apparent visible difference Scheller observed between wildtype and either allele of mutants at any developmental stages. However, both alleles of *rwa2* have displayed increased resistance toward *Botrytis cinerea*, a necrotrophic fungus. The other mutants, *rwa1*, *rwa3*, and *rwa4*, did not have detectable changes in acetylation, presumably due to genetic redundancy. Double, triple and quadruple mutants are currently being investigated.

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