

## **Functional and structural characterization of rice cellulose synthase-like f6 loss-of-function mutants**

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### **Project Goals:**

#### **Identify cell wall changes associated with lack of mixed linkage glucan accumulation in grasses**

Mixed-linkage glucan (MLG) is a cell wall polysaccharide containing a backbone of unbranched (1,3)- and (1,4)  $\beta$ -glucosyl residues. Based on its occurrence in plants and chemical characteristics, MLG has primarily been associated with the regulation of cell wall expansion due to its high and transient accumulation in young, expanding tissues. The Cellulose synthase-like F (CslF) subfamily of glycosyltransferases has previously been implicated in mediating the biosynthesis of this polymer. We have confirmed that the rice (*Oryza sativa*) CslF6 gene mediates the biosynthesis of MLG by overexpressing it in *Nicotiana benthamiana*. Rice cslf6 knockout mutants display a drastic decrease in MLG content (97% reduction in coleoptiles and virtually undetectable in other tissues) but otherwise grew normally during vegetative development, showing only a moderate decrease in both plant height and stem diameter. These results challenge previous assumptions on the role of MLG in grass cell wall structure and function. We present here preliminary results, using primarily Fourier Transform Mid-Infrared spectroscopy, that indicate that significant changes occur in cslf6 mutant cell walls in specific tissue and cell types of roots and coleoptiles. These results highlight possible structural modifications in plant polysaccharide organization that occur in response to the loss of MLG in grass cell walls.

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