Identification and validation of a key gene controlling differential flowering time between switchgrass ecotypes

Soyeon Choi*[1,3] (Soyeon.Choi@uga.edu), Ratul Chowdhury[2,3], Peng Qi[1,3], Tom H. Pendergast IV[1,3], Katrien M. Devos[1,3], and **Gerald A. Tuskan**[3]

[1]University of Georgia, Athens, GA; [2]The Pennsylvania State University, University Park, PA; [3]Center for Bioenergy Innovation, Oak Ridge National Laboratory, TN.

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Project Goals: The Center for Bioenergy Innovation (CBI) vision is *to accelerate domestication of bioenergy-relevant, non-model plants and microbes to enable high-impact innovations at multiple points in the bioenergy supply chain.* CBI addresses strategic barriers to the current bioeconomy in the areas of 1) high-yielding, robust feedstocks, 2) lower capital and processing costs via consolidated bioprocessing (CBP) to specialty biofuels, and 3) methods to create valuable byproducts from the lignin. CBI will identify and utilize key plant genes for growth, composition, and sustainability phenotypes as a means of achieving lower feedstock costs, focusing on poplar and switchgrass. We will convert these feedstocks to biofuels using CBP with cotreatment at high rates, titers and yield in combination with catalytic upgrading into drop-in hydrocarbon fuel blendstocks.

Flowering time in switchgrass, a crucial determinant of biomass yield, differs between upland and lowland ecotypes. Lowland switchgrass flowers later than uplands but key genes contributing to the delayed flowering in lowlands have not been elucidated. In this research, *PvHd1,* orthologous to rice *Hd1,* has been identified and validated as a causal gene to the differential flowering time between switchgrass ecotypes. This provides the first example in switchgrass of QTL data advancing to validated underlying gene. Two alleles of *PvHd1* encoded proteins that differed by an ecotype-specific amino acid in B-Box domain 1 and the protein variants were predicted to be folded differentially due to the single amino acid substitution (p.S35G). The effect of the amino acid variant was assessed in *CONSTANS* (*CO*, orthologous to rice *Hd1*)-null *Arabidopsis* plants, showing that the p.S35G substitution is causal to the delayed flowering time. The findings from this research will help control the onset of flowering in switchgrass and contribute to acquiring higher biomass from the bioenergy crop across a wider cultivation area.

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