Characterization and Stress Response of Monolignol *p*-Hydroxybenzoyltransferase in Poplar

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Project Goals: Establish the scientific knowledge and new technologies to transform the maximum amount of carbon available in bioenergy crops into biofuels and bioproducts.

Lignin in many species including *Populus* spp. is decorated with *p*-hydroxybenzoate. However, the molecular basis for such structural modification remains largely undetermined. Here, we report the identification and characterization of a *Populus* BAHD family acyltransferase that catalyzes monolignol *p*-hydroxybenzoylation, thus controlling the formation of *p*-hydroxybenzoylated lignin structures. With *in vitro* systematic screening and biochemic characterization of *Populus* BAHD family acyltransferases, we reveal that a *Populus* acyltransferase PHBMT1 kinetically preferentially uses *p*-hydroxybenzoyl-CoA to acylate syringyl lignin monomer sinapyl alcohol *in vitro*. Consistently, disrupting *PHBMT1* in *Populus* via CRISPR–Cas9 gene editing nearly completely depletes *p*-hydroxybenzoates of stem lignin; conversely, overexpression of *PHBMT1* enhances stem lignin *p*-hydroxybenzoylation, suggesting that PHBMT1 functions as a prime monolignol *p*-hydroxybenzoyltransferase *in planta*. Altering lignin *p*-hydroxybenzoate modification substantially changes the lignin solvent dissolution rate, indicative of its structural significance on lignin physicochemical properties. *PHBMT1* is highly induced with mechanical stress or gravistimulation along with the tension wood formation. Altering lignin *p*-hydroxybenzoylation changes plant's autotropism and gravitropism behaviors. Identification of monolignol *p*-hydroxybenzoyltransferase offers a valuable tool for tailoring lignin structure and physicochemical properties and for engineering the industrially important platform chemical in woody biomass.

Publications

1. Zhao, Y., *et al.* (2021) Monolignol acyltransferase for lignin *p*-hydroxybenzoylation in *Populus*. *Nature plants* 7, 1288-1300

2. Zhao, Y., *et al.* (2021) The Inducible Accumulation of Cell Wall-Bound *p*-Hydroxybenzoates Is Involved in the Regulation of Gravitropic Response of Poplar. *Front Plant Sci* 12, 755576

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