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Design of Transgenic *S. cerevisiae* for Enzymatic Pretreatment

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ABSTRACT

Biofuels, combustible fuel produced from fermentation of agricultural biomass by microorganisms, represent one of the best possible paths forward for sustainable energy production. However, inefficiencies in biofuel production create barriers that stand in the way of their widespread adoption. One such barrier is the breakdown of lignin, a biopolymer that exists on the edge of plant cell walls which protects the sugars that are used in fermentation. Currently, lignin is broken down in energy-intensive thermal pretreatment processes. A viable alternative is the expression of lignin-degrading enzymes by synthetic microorganisms that work at standard temperatures, eliminating the need for the high-energy input of thermal pretreatment. Four lignin-degrading enzymes were selected from termites (*R. flavipes*) and white rot fungus (*C. fiorinae* PJ7) and two helper enzymes that assist in lignin degradation were selected and then optimized for expression in yeast. The genetic devices amplified were assembled using standard DNA assembly methods. Future transformation into yeast (*S. cerevisiae*) cells and testing of lignin-breakdown effectiveness may open up an alternative path for thermal pretreatment of biomass.

KEYWORDS

biofuels, lignin, enzymatic pretreatment, biomass, *S. cerevisiae*