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Co-expression of *TAL1* and *ADH1* in recombinant xylose-fermenting *Saccharomyces cerevisiae* improves ethanol production from lignocellulosic hydrolysates in the presence of furfural

Abstract

Lignocellulosic biomass dedicated to bioethanol production usually contains pentoses and inhibitory compounds such as furfural that are not well tolerated by *Saccharomyces cerevisiae*. Thus, *S. cerevisiae* strains with the capability of utilizing both glucose and xylose in the presence of inhibitors such as furfural are very important in industrial ethanol production. Under the synergistic conditions of transaldolase (TAL) and alcohol dehydrogenase (ADH) overexpression, *S. cerevisiae* MT8-1X/TAL–ADH was able to produce 1.3-fold and 2.3-fold more ethanol in the presence of 70 mM furfural than a TAL-expressing strain and a control strain, respectively. We also tested the strains' ability by mimicking industrial ethanol production from hemicellulosic hydrolysate containing fermentation inhibitors, and ethanol production was further improved by 16% when using MT8-1X/TAL–ADH compared to the control strain. Transcript analysis further revealed that besides the pentose phosphate pathway genes *TKL1* and *TAL1*, *ADH7* was also upregulated in response to furfural stress, which resulted in higher ethanol production compared to the TAL-expressing strain. The improved capability of our modified strain was based on its capacity to more quickly reduce furfural in situ resulting in higher ethanol production. The co-expression of *TAL/ADH* genes is one crucial strategy to fully utilize undetoxified lignocellulosic hydrolysate, leading to cost-competitive ethanol production.

Keywords: Furfural, *TAL1*, *ADH1*, Overexpression, Hemicellulosic hydrolysate, Xylose, *Saccharomyces cerevisiae*, Bioethanol