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Poster Abstract Funding Area: Biosystem Design
Abstract file naming convention: Zhao_**Yoshikuni**.pdf

Evaluation of *Issatchenkia orientalis* as a next-generation chassis for bioproduct production from lignocellulosic biomass

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Project Goals: We aim to characterize *Issatchenkia orientalis* as a chassis to produce bioproducts directly from lignocellulosic hydrolysates and identify benchmark strains for metabolic engineering.

Abstract

During pretreatment and hydrolysis of lignocellulosic biomass, many inhibitors are generated as by-products, including organic acids, furan derivatives, and phenolic compounds. These inhibitors negatively affect microbial growth and bioproduct production from lignocellulosic sugars. Conventional industrially relevant (model) microbes such as *E. coli* and *S. cerevisiae* cannot tolerate these inhibitors. *I. orientalis* is a non-model ascomycetes yeast species distinguished by its ability to tolerate multiple stresses. This species may be a promising alternative for producing bioproducts from lignocellulosic substrates. To characterize this species and identify benchmark strains, we collected 160 *I. orientalis* strains from culture collections globally. We are currently evaluating their abilities to tolerate pH and temperature fluctuations and various lignocellulosic inhibitors (e.g., HMF, furfural, acetate, NaCl, and phenolics), as well as their glycolytic capacity, substrate range (e.g., growth on xylose, arabinose, and cellobiose), growth rate, and nutritional requirements (e.g., vitamins and amino acids). The *I. orientalis* strains evaluated thus far generally show promising ability to tolerate low (3.0) pH and various lignocellulosic inhibitors, as well as to produce ethanol from glucose at near quantitative (100%) yield. These strains were more robust when fermentation conditions were semi-aerobic than when they were aerobic, suggesting that that strains use oxygen to reduce inhibitor activity . Most of the strains could catabolize arabinose and cellobiose as carbon sources when glucose was depleted, but they could rarely use xylose.

Funding statement: *This work was funded by the DOE Center for Advanced Bioenergy and Bioproducts Innovation (U.S. Department of Energy, Office of Science, Office of Biological and Environmental Research under Award Number DE-SCxxxxx). Any opinions, findings, and conclusions or recommendations expressed in this publication are those of the authors and do not necessarily reflect the views of the U.S. Department of Energy.*