

LIPID PRODUCTION FROM BIOMASS VIA OLEAGINOUS YEAST

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Oleaginous microbes have received significant attention for the production of renewable diesel given their innate capacity to produce high cellular density of lipids, typically under nutrient deprivation, from lignocellulose-derived sugars. To date, significant work has been conducted on several model oleaginous microbes, mostly on clean feedstocks. To produce renewable diesel production from lignocellulosic sugars, it is essential to screen potential host organisms on realistic biomass-derived hydrolysates. To that end, here we report shake flask screening of 33 oleaginous yeasts, which were down-selected based on lipid titer. From there, 9 oleaginous yeast strains were carried forward for several iterations of batch fermentation, using two substrates: glucose, and a mixed sugars stream representative of biomass hydrolysates. Based on fermentation metrics, three strains were selected to assess their performance on hydrolysate from a pilot-scale pretreatment and enzymatic hydrolysis of corn stover. The fermentation results on biomass hydrolysate showed that *Cryptococcus curvatus* exhibits the highest overall productivity ($0.10 \text{ g L}^{-1} \text{ h}^{-1}$), *Rhodospiridium toruloides* the highest content (67 % FAME), and *Trichosporon guehoae* the highest yield (0.29 g g^{-1}). We also demonstrate the mechanisms that oleaginous yeast employ to detoxify hydrolysate including utilization of lignin-derived aromatic compounds, in addition to lipid and sterol speciation for the selected strains. Data on lipid recovery, using a mild acid treatment and extraction approach, and hydrotreating the recovered lipids to green diesel, are also presented. Taken together, these results demonstrate the potential for an integrated process for renewable diesel production, identify several oleaginous strains for further development, and highlight opportunities for improvements to an oleaginous microbial platform for the production of renewable, drop-in replacement diesel.