

Optimization of Pre-commercial Enzymes Dosage for a Potential Lignocellulosic Biorefinery

Background/Objective

Lignocellulolytic enzymes are a primary cost constraint in second-generation ethanol biorefineries, accounting for up to 30% of operating expenses. Due to limited inputs and achieving industrially viable ethanol titers and yields with an inexpensive nitrogen source, the hydrothermal pretreatment approach is a leading biorefinery technique. However, enzyme loadings reported in the literature vary widely, and optimization among diverse bioenergy feedstocks has not been systematically investigated, hindering cost assessments. We assessed enzyme dosages (ED) of cellulase and hemicellulase on hydrothermally pretreated lignocellulosic substrates to maximize sugar and ethanol yields and improve cost analysis.

Approach

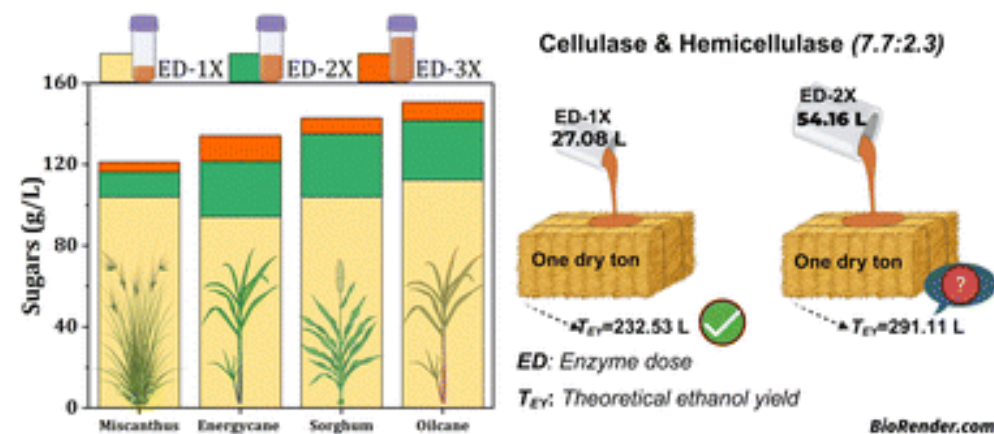
Three ED levels of cellulase (NS22257):hemicellulase (NS22244) combinations, 20 mg:7.25 mg (ED-1), 40 mg:14.5 mg (ED-2), and 60 mg:21.75 mg (ED-3), were evaluated in fed-batch hydrolysis of hydrothermally pretreated lignocellulosic substrates of oilcane, energycane, sorghum, and miscanthus feedstocks. The optimum ED condition was scaled up in a 75 L bioreactor to assess industrial feasibility. Additionally, a semi-integrated bioprocess was implemented to improve ethanol yield using engineered *Saccharomyces cerevisiae*.

Results

As expected, the highest sugar yields were obtained with the highest ED (ED-3), but the difference between ED-2 and ED-3 was not substantial for the selected feedstocks. While all EDs enabled sufficient sugar release to achieve industrial ethanol titers (≥ 40 g/L), ED-2 was optimal for sugar release, whereas ED-1 was optimal for ethanol yield (213 L) at a tenfold lower enzyme dosage than reported in prior studies.

Significance/Impacts

Optimized enzyme dosages could improve the overall bioprocess economics of lignocellulosic biorefineries.



A clear relationship emerged between sugar concentrations, enzyme dosage, and ethanol yield per ton of biomass.