Engineering Multi-Enzyme Whole-Cell Biocatalysts for Biofuel Production

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In this project, I successfully studied the relationship between multi-enzyme biocatalyst structure and performance and developed a comprehensive quantitative model to predict the assembly efficiency of primary scaffold proteins (pScafs) to anchor scaffold proteins (aScaf), the overall assembly efficiency of multi-scaffolded enzyme assemblies (mSEAs) on yeast cell surfaces to achieve the optimal enzyme density as well as the performance of mSEAs in the direct conversion of cellulose to ethanol. Experimental trials were performed based on a partial factorial Design of Experiments (DOE) of three design variables. The three design variables are aScaf complexity at three levels (aScaf1, aScaf2, and aScaf3), aScaf display level on yeast cell surfaces at nine levels (aScaf-DL1 to aScaf-DL9), and ten combinations of four cellulase enzymes (BGL, EG, CBHI, and CBHII) assembled on the pScafs. The model and the statistical analysis revealed some critical insights on how the molecular crowding on the yeast-cell surface limits the assembly efficiency of mSEAs and the resulting enzyme density. The study demonstrated the power of a quantitative model in guiding the rational design and engineering of multi-enzyme whole-cell biocatalysts to achieve optimal assembly efficiency for direct conversion of biomass to ethanol. In the future, these surface engineered yeast cells can be applied in the consolidated bioprocessing (CBP) to realize the biomass degradation and ethanol production in a single step, which is critical for developing a sustainable biomass-based refinery.

Awards Won:

NC State College of Engineering: Award to attend NC State Engineering Summer Camp

First Award of $5,000