

Application of a Designed Protein Cage to Increase Pathway Flux of Cellulolysis

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Project Goals: The goal of this work is to improve methods for breaking down cellulose to glucose in the laboratory by using designed protein cages as a scaffold for cellulase enzymes. This technology would aid in the development of efficient routes to production of ethanol and other biocommodities from microbial sources.

Motivated by the rich diversity of protein molecules that have evolved in nature to form complex and highly symmetric supramolecular structures, recent engineering efforts in the field of protein design have exploited symmetry to create novel self-assembling protein structures of types unseen in biology. Advances in designing proteins to self-assemble into specific architectures are now opening up numerous exciting technology applications. One such application of protein cages is in enzyme display for improving pathway flux of sequentially acting enzymes. We have created protein cages with cellulolytic activity by using the sortase enzyme SrtA to covalently link multiple cellulase enzymes to the exterior of a designed protein cage. By decorating these cages with both the endoglucanase Cel8A and the exoglucanase Cel48S, we have demonstrated a more than 5-fold increase in pathway flux of cellulolysis compared to the free enzymes in solution. These cellulolytic cages could find application in engineering more efficient routes to the production of ethanol and other biocommodities.

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