

## **Functional assembly of mini-cellulosomes by synthetic yeast consortia toward one-step cellulosic ethanol production**

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Lignocellulose is the most abundant renewable natural resource for conversion to ethanol. The rate-limiting step in conversion of cellulose to fuels is the hydrolysis, especially the initial attack on these insoluble substrates. Cellulosome, an enzymatic complex usually found in anaerobic cellulolytic microorganisms, is a promising tool towards the goal of consolidated bioprocessing (CBP) for biofuel production due to its synergistic activity. The recruitment of dockerin-tagged enzymatic subunits onto cohesion-based scaffoldin modules directs the tenacious architecture of these complex cellulosomes. However, due to the high affinity of cohesin-dockerin interaction, attempts to assemble all these components have not yet been successful due to its heavy metabolic burden and potential blocking of the secretion machinery. To solve these problems, a synthetic yeast consortium capable of secreting dockerin-borne cellulases, including endoglucanase, exoglucanase, and glucosidase, for their self assembly onto cells displaying scaffoldins was constructed for the functional presentation of complex cellulosome structures. This synthetic consortium was able to functionally assemble a mini-cellulosome on the yeast surface, synergistically hydrolyze cellulose and ferment the resulting sugars to ethanol at near theoretical yield.