Paper

Insights into the mechanisms of synergistic interactions between auxiliary activity (AA) family 9 enzymes and cellulases during biomass deconstruction

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Enzyme based “biorefinery” processes continue to aspire to achieve faster, more complete hydrolysis of the cellulosic component of biomass to monomeric sugars (which can be subsequently converted to a wide range of fuels/chemicals/biomaterials). Typically, too high enzyme/protein loading is still required to achieve high sugar yields. More recently, lytic polysaccharide monooxygenases (LPMOs) such as AA9, have been shown to interact synergistically with cellulases to enhance the enzymatic hydrolysis of various “commercially-relevant” pretreated and “model” cellulosic substrates. In most cases, the addition of LPMOs consequently reduced enzyme/protein loading required to achieve effective cellulose hydrolysis. While good progress has been made in better defining protein structure and catalytic mechanism of AA9, the way in which this enzyme enhances hydrolytic potential of a “cellulase mixture” during hydrolysis of cellulosic substrates is still not completely understood. The presentation will describe the synergistic cooperation between AA9 and cellulases during hydrolysis of various model and pretreated substrates. The influence that the physicochemical characteristics of the substrates have on the ability of AA9 to enhance the hydrolytic potential of cellulase mixtures will be elucidated. A double-antibody sandwich enzyme linked immunosorbent assay (ELISA) was used to monitor the specific cellulase monocomponents and AA9 adsorption/desorption profiles during cellulose hydrolysis. The addition of AA9 increased the desorption of Cel7A (a processive exoglucanase), but not Cel7B (an endoglucanase). It was apparent that the relative amount of accessible crystalline cellulose, as assessed using a specific cellulose binding module (CBM) based probe, was the substrate characteristic that most influenced AA9/cellulase synergy.