World Academy of Science, Engineering and Technology
International Journal of Energy and Environmental Engineering
Vol:11, No:12, 2017

Heterologous Expression of a Clostridium thermocellum Proteins and Assembly of Cellulosomes 'in vitro' for Biotechnology Applications

Authors : Jessica Pinheiro Silva, Brenda Rabello De Camargo, Daniel Gusmao De Morais, Eliane Ferreira Noronha

Abstract : The utilization of lignocellulosic biomass as source of polysaccharides for industrial applications requires an arsenal of enzymes with different mode of action able to hydrolyze its complex and recalcitrant structure. Clostridium thermocellum is gram-positive, thermophilic bacterium producing lignocellulosic hydrolyzing enzymes in the form of multi-enzyme complex, termed celulossomes. This complex has several hydrolytic enzymes attached to a large and enzymically inactive protein known as Cellulosome-integrating protein (CipA), which serves as a scaffolding protein for the complex produced. This attachment occurs through specific interactions between cohesin modules of CipA and dockerin modules in enzymes. The present work aims to construct celulosomes in vitro with the structural protein CipA, a xylanase called Xyn10D and a cellulose called CelJ from C.thermocellum. A mini-scafoldin was constructed from modules derived from CipA containing two cohesion modules. This was cloned and expressed in Escherichia coli. The other two genes were cloned under the control of the alcohol oxidase 1 promoter (AOX1) in the vector pPIC9 and integrated into the genome of the methylotrophic yeast Pichia pastoris GS115. Purification of each protein is being carried out. Further studies regarding enzymatic activity of the cellulosome is going to be evaluated. The cellulosome built in vitro and composed of mini-CipA, CelJ and Xyn10D, can be very interesting for application in industrial processes involving the degradation of plant biomass.

Keywords : cellulosome, CipA, Clostridium thermocellum, cohesin, dockerin, yeast
Conference Title : ICBB 2017 : International Conference on Biofuels and Bioenergy
Conference Location : Paris, France
Conference Dates : December 28-29, 2017