

CLONING, PURIFICATION, AND BIOCHEMICAL CHARACTERIZATION OF AN ESTERASE FROM *ASPERGILLUS NIDULANS*

Molecular Biotechnology, Systems Biology and Metabolic Engineering

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Body

A large accumulation of agro-industrial waste from different segments is generated daily and is often not properly managed. There are now other fronts in research to give a destination to these residues; these studies are generally aimed at obtaining new and better enzymes and the formulation of enzymatic cocktails that contain (for example, cellulases and hemicellulases) responsible for the degradation of lignocellulosic material. The plant cell wall is mainly composed of cellulose, hemicellulose, and lignin, forming a complex structure. Xylan is one of the main constituents of hemicellulose. To degrade this structure, enzymatic hydrolysis must occur synergistically with xylanolytic enzymes, such as endo-beta-1,4-xylanases, β -xylosidases, and acetyl xylan esterase (AXE). In the current work, we reported the purification and biochemical characterization of an acetyl xylan esterase (AxeCE3) from *Aspergillus nidulans*. The *axeCE3* gene was cloned into the pEXPYR vector and transformed into *A. nidulans* A773 for protein expression. The enzyme AxeCE3 was purified and characterized for its biochemical properties. AxeCE3 showed activity over a wide range of pH (3.0-9.0) and temperature (30-70 °C), with maximum activity at 55 °C, pH 7.0. Regarding the stability at temperature, AxeCE3 showed values above 90% of residual activity after 24 h of incubation at 45 and 50 °C. In relation to stability at pH, AxeCE3 maintained more than 90% of its residual activity after being incubated at 25 °C for 24 h between the pH range 3.0 to 9.0. It was also verified the effect of possible inhibitors (ethylenediamine tetraacetic acid (EDTA), Furfural, and 5-Hydroxymethylfurfural (5-HMF)) on the enzyme activity. AxeCE3 maintained 88% of relative activity at 5 mM EDTA, 43% and 82% at 50 mM furfural and 5-HMF, respectively. The results showed that AxeCE3 has interesting properties to use in the development in the formulation of enzymatic cocktails for the hydrolysis of lignocellulosic residues.

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Palavras-chave : acetyl xylan esterase, *Aspergillus nidulans*, lignocellulosic residues