Kharazmi University
Faculty of Science - Biological Science Department
Biochemistry Devision

Thesis Title:

Partial Biochemical characterization of b-glucanase enzymes isolated from native thermophilic bacteria Cohnella sp.A01

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Abstract

Non cellulytic beta glucanases which in most cases synthesized during plant invasion by some plants and Mycoparasitic fungi, hydrolyzed cell walls of pathogenic fungi and yeasts to protect of plants. The aim of this study was to investigate the biochemical properties of the beta glucanase from thermophilic native bacteria, *Cohnella* sp.A01.In line with this objective, the cloning of beta glucanases gene and its expression in *E.coli* was performed. In this study, the lipase gene from the bacterium *Cohnella* sp.A01was cloned in to pET26b(+) vector and expressed in *E.coli*(DE3). The expressed protein bears 639 amino acids with sequence homology to glycosyl hydrolases family 30 (GH30). The enzyme was purified using Ni-NTA Column. Laminarin and pustulan as substrarte to measure enzyme activity, were selected. The biochemical properties of the enzyme, including the effect of pH and temperature, as well as detergents and metal ions were measured. In his study, beta 1,6-glucanase gene was successfully cloned and expressed in *E.coli*.The enzyme molecular weight is 64 kDa and purification efficiency was 46%. The enzyme has a specific activity 8.71 U/mg on laminarin and 5.47 U/mg on pustulan. The enzyme was identified as a beta 1,6-glucanase. The optimum temperature and pH for the enzyme were 50C and 8 respectively. The enzyme has good stability at high temperature and in the range of pH 7 to 9. Enzyme activity in the presence of metal ions and chemicals used, did not rise. Urea, SDS and copper metal ions had the highest inhibitory effect on the enzyme activity.

Key words: Beta 1,6 glucanase- *Cohnella* sp.A01-biochemical properties- *E.coli*