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Production, Purification and Characterization of Inulinase from a Newly Isolated *Streptomyces* sp. CP01

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ABSTRACT

Inulinase is an enzyme catalyzing the hydrolysis of inulin, a plant reserve polysaccharide, into fructoses and fructooligosaccharides which are widely used as food additives. Here we report inulinase from a newly isolated *Streptomyces* as in the past decade there have been very few reports on inulinases from *Streptomyces*, especially purification and characterization of these enzymes. Out of 371 *Streptomyces* isolates, *Streptomyces* sp. CP01 produced highest inulinase activity of 0.50 U/ml. The enzyme activity was increased to 1.60 U/ml when CP01 was cultivated under the optimal conditions which consisted of using basal medium (Czapek's Dox) containing 1% (w/v) inulin extract from Jerusalem artichoke's root tubers and 0.7% (w/v) tryptone at pH8, shaking at 200 rpm and 28℃ for 24 h. The enzyme was purified from culture filtrate to about 67-fold purity by $(NH_4)_2SO_4$ precipitation followed by four consecutive column chromatography steps. The purified enzyme is a single peptide with approximate molecular mass of 73 kDa as analyzed by gel filtration and 70.8 kDa as assessed by SDS-PAGE. The enzyme is optimally active at 55℃ and pH 6.0, however it still possesses more than 80% of the maximal activity at pH ranging from 5.5 to 9.0. It is stable at temperature up to 50℃ and at broad range of pH from 5.0 to 9.0 for 30 min. Its K_m and V_{max} values for inulin were 2.34 mM and 440 μmol·min^{-1}·mg^{-1}, respectively. This enzyme has potential for industrial application as it is active at moderately high temperature and wide range of pH.

KEYWORDS

Characterization; Inulinase; Production; Purification; *Streptomyces*

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