World Academy of Science, Engineering and Technology
International Journal of Agricultural and Biosystems Engineering
Vol:10, No:01, 2016

Production and Purification of Pectinase by Aspergillus Niger

Authors : M. Umar Dahot, G. S. Mangrio

Abstract : In this study Agro-industrial waste was used as a carbon source, which is a low cost substrate. Along with this, various sugars and molasses of 2.5% and 5% were investigated as substrate/carbon source for the growth of A.niger and Pectinase production. Different nitrogen sources were also used. An overview of results obtained show that 5% sucrose, 5% molasses and 0.4% (NH4)2SO4 were found the best carbon and nitrogen sources for the production of pectinase by A. niger. The maximum production of pectinase (26.87units/ml) was observed at pH 6.0 after 72 hrs incubation. The optimum temperature for the maximum production of pectinase was achieved at 35ºC when maximum production of pectinase was obtained as 28.25Units/ml.Pectinase enzyme was purified with ammonium sulphate precipitation and dialyzed sample was finally applied on gel filtration chromatography (Sephadex G-100) and Ion Exchange DEAE A-50. The enzyme was purified 2.5 fold by gel chromatography on Sephadex G-100 and Four fractions were obtained, Fraction 1, 2, 4 showed single band while Fraction -3 showed multiple bands on SDS Page electrophoresis. Fraction -3 was pooled, dialyzed and separated on Sephdex A-50 and two fractions 3a and 3b showed single band. The molecular weights of the purified fractions were detected in the range of 33000 ± 2000 and 38000± 2000 Daltons. The purified enzyme was specifically most active with pure pectin, while pectin, Lemon pectin and orange peel given lower activity as compared to (control). The optimum pH and temperature for pectinase activity was found between pH 5.0 and 6.0 and 40°- 50°C, respectively. The enzyme was stable over the pH range 3.0-8.0. The thermostability of was determined and it was observed that the pectinase activity is heat stable and retains activity more than 40% when incubated at 90°C for 10 minutes. The pectinase activity of F3a and F3b was increased with different metal ions. The Pectinase activity was stimulated in the presence of CaCl2 up to 10-30%. ZnSO4, MnSO4 and Mg SO4 showed higher activity in fractions F3a and F3b, which indicates that the pectinase belongs to metalo-enzymes. It is concluded that A. niger is capable to produce pH stable and thermostable pectinase, which can be used for industrial purposes.

Keywords : pectinase, a. niger, production, purification, characterization
Conference Title : ICABBBE 2016 : International Conference on Agricultural, Biotechnology, Biological and Biosystems Engineering
Conference Location : Jeddah, Saudi Arabia
Conference Dates : January 26-27, 2016