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Characterization of Pectinase from Local Microorganisms to Support Industry Based Green Chemistry

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Abstract: Pectinase are enzymes that hydrolyze pectin compounds. The use of this enzyme is primarily to reduce the viscosity of the beverage thus simplifying the purification process. Pectinase activity influenced by microbial sources. Exploration of two types of microbes that Aspergillus spp. and Bacillus spp. pectinase give different performance, but the use of local strain is still not widely studied. The aim of this research is exploration of pectinase from A. niger and B. firmus include production conditions and characterization. Bacillus firmus incubated and shaken at a speed of 200 rpm at pH variation (5, 6, 7, 8, 9, 10), temperature (30, 35, 40, 45, 50) °C and incubation time (6, 12, 18, 24, 30, 36) hours. Media was centrifuged at 3000 rpm, pectinase enzyme activity determined. Enzyme production by A. niger determined to variations in temperature and pH were similar to B. firmus, but the variation of the incubation time was 24, 48, 72, 96, 120 hours. Pectinase crude extract was further purified by precipitation using ammonium sulfate saturation in fraction 0-20 %, 20-40 %, 40-60 %, 60-80 %, then dialyzed. Determination of optimum conditions pectinase activity performed by measuring the variation of enzyme activity on pH (4, 6, 7, 8, 10), temperature (30, 35, 40, 45, 50) °C, and the incubation time (10, 20, 30, 40, 50) minutes. Determination of kinetic parameters of pectinase enzyme reaction carried out by measuring the rate of enzyme reactions at the optimum conditions, but the variation of the concentration of substrate (pectin 0.1% , 0.2% , 0.3% , 0.4% , 0.5%). The results showed that the optimum conditions of production of pectinase from B. firmus achieved at pH 7-8.0, 40-50 °C temperature and fermentation time 18 hours. Purification of pectinase showed the highest purity in the 40-80 % ammonium sulfate fraction. Character pectinase obtained: the optimum working conditions of A. niger pectinase at pH 5, while pectinase from B. firmus at pH 7, temperature and optimum incubation time showed the same value, namely the temperature of 50 °C and incubation time of 30 minutes. The presence of metal ions can affect the activity of pectinase, the concentration of Zn 2 +, Pb 2 +, Ca 2 + and K + and 2 mM Mg 2 + above 6 mM inhibit the activity of pectinase.

Keywords: pectinase, Bacillus firmus, Aspergillus niger, green chemistry

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