E2-36: Kinetic studies of extracellular α-galactosidase from *Citrobacter ferundii*

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α-galactosidase is used in soyamilk processing, production of gelling agents, pulp and paper industry and beet sugar production. In industrial processes the enzymes used are mainly extracellular bacterial enzymes that are stable at room temperature. In this study, extracellular α-galactosidase producing bacteria was isolated from soil, using a raffinose culture medium. The species that produced the highest α- galactosidase activity of 19 milliunits/ml of culture medium was identified as *Citrobacter ferundii* by morphological and biochemical tests.

α-galactosidase from *Citrobacter ferundii* was purified by ammonium sulphate fractionation and DEAE ion exchange chromatography. A 164 fold purification was obtained with a yield of 8%. Polyacrylamide gel electrophoresis of the enzyme showed 2 protein bands. In kinetic studies of the partially purified enzyme, p-nitro phenyl α-D-galactopyranoside was used as the substrate. The Michaelis constant and maximum reaction velocity obtained were 2.85×10^{-3}M and 14 μmol/min/mg protein. Studies on the effect of pH on enzyme activity showed a broad pH optimum from 5.5 to 8.0 with maximum activity at pH 7.5. The enzyme was stable between pH 5.5 to 8.0. The optimum enzyme activity was observed at 40°C. The enzyme was stable upto 40°C. The enzyme solution did not contain any invertase activity.