

SUMMARY

Two α Galactosidase isoenzymes have been isolated from coconut kernel and characterized using sephadex column chromatography. The elution had been done using Mcilvaine buffer (pH 5.5) containing KCl (Balasubramaniam et al 1974). The purity of these enzymes have not been tested. In this study α Galactosidase was eluted from the sephadex column using Mcilvaine buffer (pH 5.5) without KCl. All the enzyme was eluted in the low molecular isoenzyme form. Recovery from the column was 23.6 percent of the original extract, the purity of the enzyme increasing 171 fold. The enzyme thus obtained was further purified by concentration using ultrafiltration and ultracentrifugation. At this stage 11.5 percent of the original extract was recovered while the purity increased upto 512 fold. This enzyme preparation when tested by polyacrylamide gel electrophoresis showed a single protein band which corresponded to the fluorescent band obtained by the action of α Galactosidase on umbelliferyl α D galactopyranoside. On cellulose acetate paper electrophoresis a single enzyme activity band was obtained. As the protein concentration was too low, the corresponding protein band could not be detected. Molecular weight of the purified enzyme was 18,530. The enzyme was inhibited by a substrate concentration of para nitrophenyl α D galactopyranoside of 6×10^{-4} M. For the above substrate Km was 2.5×10^{-4} M and Vmax was 25 μ Mol/min/mg. of protein. The enzyme is stable upto 45°C. Activation energy was 6.83 Kcal per mole. Enzyme activity showed a broad pH optimum from 2.5 to 6 with a peak at pH 5.5. Enzyme was noncompetitively inhibited by mercuric ions and was competitively inhibited by D galactose.