'Studies of Invertases from the flowers of; *Woodfordia fruticosa* (Malitta) and *Madhuca longifolia* (Mi)'

A THESIS PRESENTED

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ABSTRACT

Invertase of *Woodfordia fruticosa* and *Madhuca longifolia* which were used to hydrolyze sucrose during the manufacture of Arishta (a fermented Ayurvedic drug) was purified by ammonium sulphate fractionation, sepharose 4B gel flirtation and DEAE cellulose ion exchange chromatography. Invertase of *Woodfordia fruticosa* was partially purified to near homogeneity as judged by PAGE with 17.9 % recovery whereas invertase of *Madhuca longifolia* was partially purified 52 fold with a 20.4 % recovery.

Molecular masses of the purified *Woodfordia* and *Madhuca* invetases as determined by sepharose 4B gel filtration were ~ 230 kDa and ~ 210 kDa respectively. Faint bands seen at 67 kDa and 43 kDa and at 43 kDa and 14 kDa in SDS-PAGE of *Woodfordia* and *Madhuca* invertases respectively suggest that possibly these two enzymes would be isozymes.

Both enzymes showed a broad pH optimum between 4-7 and maximum activity at 37 °C. Above 45 °C activity of both enzymes slowly declined and inactivated at 80 °C. Approximate Km values of *Woodfordia* and *Madhuca* invertases for sucrose were 160 mM and 125 mM respectively.

Fructose was a competitive inhibitor, whereas mercury, urea, manganese, semicarbazide hydrochloride, zinc and cadmium were mixed inhibitors for the invertase of *Woodfordia*.

Manganese , mercury and semicarbazide hydrochloride were non-competitive inhibitors, zinc was a competitive inhibitor and fructose, cadmium and urea were mixed inhibitors for the invertase of *Madhuca*.

Tyrosine and valine acted as activators for both enzymes and between these two, the effect of tyrosine is significant.