

**ISOLATION AND CHARACTERIZATION OF  
INVERTASE FROM *SACCHAROMYCES CEREVISIAE*  
OF LOCAL TODDY JUICE**

**Ph.D.DISSERTATION**

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**Ph.D. ( Chemistry )**

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**Title** : Isolation and Characterization of Invertase from *Saccharomyces cerevisiae* of Local Toddy Juice

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**Abstract** : The main aim of this work was to investigate the isolation and characterization of invertase from *Saccharomyces cerevisiae*. The yeast strain (*S.cerevisiae*) was isolated from toddy juice by dilution-plate method and studied under microscope. Malt extract agar medium was used for the cultivation of yeast. The yeast was identified by sugar fermentation test, sugar assimilation test, nitrate assimilation test and ethanol as sole source of carbon. The effect of different fermentation periods of isolated yeast had been studied to obtain the optimum period. Invertase ( E.C. 3.2.1.26 ) was purified from crude extract by two steps chromatography, viz., Sephadex G-150 gel filtration and DEAE Sephadex ion exchange chromatography. The specific activity of invertase was increased by 2.13 fold over that of crude extract. This enzyme was confirmed by non SDS-PAGE as single bands. After non SDS-PAGE, the molecular weight for invertase was 269,000. The invertase from *S.cerevisiae* had optimum

temperature value of 55° C at optimum pH of 4.5. The  $K_m$  and  $V_{max}$  values of the purified invertase were found to be 14.3651 mM and 0.2186 mMmin<sup>-1</sup>. The above studies were carried out using UV-visible spectrophotometry. The enzyme invertase was immobilized with alginate. The hydrolysis of sucrose with invertase was confirmed with Benedict's reagent.