

عنوان مقاله:

Isolation, Purification and Partial Characterization of Intracellular Invertase from Palm Wine Yeast (*Saccharomyces* sp.)

محل انتشار:

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خلاصه مقاله:

Introduction: Invertase belongs to the class of enzymes called glycosidase. The enzyme is responsible for the catalytic hydrolysis of sucrose to release monosaccharides known as invert sugars. The aim of this study was the isolation, purification, and physicochemical properties of intracellular invertase from palm wine yeast (*Saccharomyces* sp.) as an alternative enzyme in several industrial applications. **Materials and Methods:** The yeast was harvested from the palm wine through flocculation and the intracellular invertase was isolated from the yeast cell by mechanical grinding using acid washed sand. The intracellular invertase was purified using combination of ion-exchange chromatography on DEAE-trisacryl and gel filtration on Sephacryl S-۳۰۰. The kinetics and other physicochemical properties of the purified enzyme were determined. **Results:** The two-step purification scheme employed gave a final yield of ۱۶۸% and a purification fold of ۳.۰. Denaturing polyacrylamide gel electrophoresis (SDS-PAGE) of the intracellular invertase gave six subunits with the molecular mass of ۷۳.۵ ± ۲.۱ kDa, ۵۲.۳ ± ۸.۱ kDa, ۴۸.۶ ± ۳.۰ kDa, ۳۷.۴ ± ۴.۸ kDa, ۲۶.۵ ± ۳.۶ , and ۲۴ ± ۴.۳ kDa, respectively, while non-denaturing PAGE revealed the presence of single entity invertase with the molecular mass of ۲۶۷ kDa. Also, native molecular mass estimated on calibrated Sephacryl S-۳۰۰ was ۲۶۶ ± ۲۸ kDa, revealing that the purified palm wine *Saccharomyces* invertase (PWSInv) is heterohexameric in nature. The K_m and V_{max} of the purified invertase were ۳۰.۸ ± ۳.۲ mM and ۹۶۷۲ ± ۱۶۹.۰ units/mg protein, respectively, leading to catalytic efficiency, k_{cat}/K_m of ۱.۴۳×۱۰^۶ M^{-۱} s^{-۱}. The optimum temperature and pH were ۶۰ °C and ۳.۰, respectively. The activation energy (E_a) of the intracellular invertase for the hydrolysis of sucrose was estimated to be ۲۸۰.۴۲ kJ/mol. **Conclusions:** The study established the presence of intracellular invertase from palm wine yeast and investigated some properties and characteristics of the purified invertase, which could be exploited for several biotechnological and industrial processes.

کلمات کلیدی:

Microbial Enzyme, Intracellular Invertase, High Fructose Syrup, *Saccharomyces* sp, Biotechnological Application

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