CONVERSION OF XYLOSE TO ETHANOL UNDER AEROBIC CONDITIONS BY CANDIDA TROPICALIS

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SUMMARY

Candida tropicalis converts xylose to ethanol under aerobic, but not anaerobic, conditions. Ethanol production lags behind growth and is accelerated by increased aeration. Adding xylose to active cultures stimulates ethanol production as does serial subculture in a medium containing xylose as a sole carbon source.

INTRODUCTION

Recent reports have shown that some yeasts will ferment the ketose sugar, xylulose, after it is formed from xylose through the action of xylose (glucose) isomerase (Wang, et al., 1980; Gong, et al., 1981). Of 42 yeasts screened for this trait, Candida tropicalis fermented xylulose at a rate exceeding that attained by any others tested, including two strains of Schizosaccharomyces pombe and five strains of Saccharomyces cerevisiae (Jeffries and Choi, 1981). Moreover, unlike S. pombe and S. cerevisiae, C. tropicalis will readily assimilate xylose. We decided, therefore, to see if aeration would enhance the fermentation of xylulose. Since we generally

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employ mixtures of xylose and xylulose to test for xylulose fermentation, we ran control cultures containing pure xylose. Ethanol was detected in these aerobic xylose cultures.

While this manuscript was in preparation, an account of ethanol production from xylose by another yeast (*Pachysolen tannophilus*) was published (Schneider *et al.*, 1981).

**MATERIALS AND METHODS**

*Candida tropicalis* ATCC 1369, *Candida* sp. ATCC 28528 and *Candida utilis* ATCC 22023 were obtained from the American Type Culture Collection, Rockville, Md. *Kluyveromyces fragilis* was obtained from J. D. Macmillan, Rutgers University, New Brunswick, N.J. All organisms were maintained on slants of yeast malt agar (Difco) at 27°C. For liquid cultivation, cells were grown in 0.67% yeast nitrogen base (YNB, Difco) plus 7.5% (w/v) xylose (Sigma, grade II). Inocula consisted of 18-hour-old cells. These were either washed in distilled water to remove contaminating glucose and other nutrients, or 1.0 ml was subcultured directly from YNB xylose medium into 32 ml of fresh medium. In either case, the initial optical density (O.D.) was 0.3 to 0.5 at 525 nm. Standard culture conditions employed 33 ml of YNB xylose medium in a 125 ml Erlenmeyer, shaken on a rotary shaker at 200 rpm (2.5 cm radius). Anaerobic conditions were similar except that flasks were fitted with sterile No. 5 rubber stoppers and flushed aseptically with N₂ for 15 minutes after inoculation or sampling. Higher aeration rates were obtained by using the same amount of medium in either 300 ml Erlenmeyer flasks or baffled 300 ml Erlenmeyer flasks at 200 or 400 rpm.

Ethanol was determined by gas chromatography on a packed glass column (60 cm x 0.2 cm) of Chromosorb 101 at 165°C, using helium as a carrier gas and a flame ionization detector. The identity of ethanol was confirmed by use of alcohol dehydrogenase (Sigma).

Xylose was determined by the method of Nelson (1944).