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Microbial Production of β-Carotene by Utilization of Agro-Industrial by-Products

By

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B.Sc. Microbiology and chemistry (2008)

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ABSTRACT

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Beta-carotene, an important fat soluble pigment, is one of the most common carotenoids and the biosynthetic precursor of vitamin A.

The current study aimed to produce beta-carotene by effective microorganism, utilizing agro-industrial by-products as cheap material. Three agro-industrial by-products, namely rice bran, molasses, sugar cane bagasse, were used for β-carotene production using three microorganisms: *Rhodotorula glutinis* ATCC 4054, *Sphingomonas paucimobilis* ATCC 10829, *Serratia marcescens* ATCC 27117 to choose the best microorganisms which produce high concentration of β-carotene.

To optimize physical and nutritional parameters for β-carotene production by *Rhodotorula glutinis* ATCC 4054 and *Serratia marcescens* ATCC 27117, using statistical approach of Response Surface Methodology (RSM) Followed by scale-up of the operation under optimized conditions.

Beta-carotene was optimized using two step approach. First, a quick identification of important factors, like carbon and nitrogen source by simple screening experiment, and second application of complex RSM for further optimization. The optimum conditions for β-carotene production by *R. glutinis* were; sucrose 18.6g/L, initial pH at 5.4, KH_2PO_4 1.01g/L and NaCl 0.66 g/L to achieve a β-carotene content from 1.23mg/kg rice bran to 3.20 mg/kg and by *S. marcescens* were Sucrose 2.5 g/L, peptone 7.8 g/L and pH at 6.7 to achieve a β-carotene content from 1.1mg/L molasses to 2.24 mg/L.

Plackett-Burman experimental design (PBD) was used for screening variables to elevated β-carotene production and Central Composite Design was proceeded to obtain main, quadratic and interaction effect of significant factors.

The produced β-carotene after scale-up under optimized conditions was purified and analyzed by Thin layer chromatography, High performance liquid chromatography and Gas chromatography-Mass spectroscopy.

The structure of beta-carotene was determined by Fourier transform infrared spectroscopy and Proton magnetic resonance. From quantitative analysis by HPLC, β-carotene had purity 68.2084% compared with standard beta-carotene and the concentration of β-carotene equal 89.06 mg/100g yeast cell.

Keywords: β-carotene, Agro-industrial by-products, *Rhodotorula glutinis*, *Sphingomonas paucimobilis*, *Serratia marcescens,* RSM, Plackett-Burman Design, Central Composite Design, TLC, HPLC, Gas chromatography-Mass spectroscopy, Fourier transform infrared spectroscopy, Proton magnetic resonance.

CONTENTS

LIST OF TABLES