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PRODUCTION OF AMYLASES BY *B.SUBTILIS* USING SUBMERGED FERMENTATION

ZAINAB REHMAN

Department of Food Technology, Institute of Foreign Trade and Management University (IFTMU), Moradabad

ABSTRACT

Eleven bacterial isolates were isolated from the three different-different places soil sample by serial dilution method and agar plate technique. These bacterial isolated colonies were identified on the basis of colony morphology. Screening of the colony for the amylase activity was done on the MAM (minimal agar medium) supplemented with 1% starch. It showed the maximal amylolytic activity and were identified as *Bacillus subtilis* in accordance to the Bergey's manual. The physical and chemical factors were also optimized. The strain was improved by UV treatment of 1, 2, 3, 4, 5 minutes. Amylase production was carried out by submerged fermentation. The amylase production was studied as function of carbon source, nitrogen source, pH and temperature. It was verified the maximum amylase activity of 0.01344U was obtained on the 4th day at 37°C and pH. Partial purification of crude amylase enzyme was done by ammonium sulphate precipitation upto 70% followed by dialysis. Pure enzyme relatively stable at wider pH range of 5-11 and the temperature 4-50°C. The activity was enhanced the present of activators like Mg²⁺. Enzyme activity was decreased in presence of inhibitors like SDS and EDTA.

Keywords: Amylase, *Bacillus subtilis*, Purification, Optimum activity, Submerged fermentation.

INTRODUCTION

Much of the history of biochemistry of enzyme research. Amylases are enzymes that breakdown starch or glycogen. The amylases can be derived from several sources such as plants, animals and microbes. Amylase belongs to group,

which is called amylolytic enzyme. Amylases are examples of hydrolases and functions in the hydrolysis of molecules. Starch exists as major carbohydrate storage product in all plants containing chlorophyll. Starch or amylum is a carbohydrate