

**STUDY ON PRODUCTION OF GLUCOSE
FROM CELLULOSIC WASTES USING
IMMOBILIZED *TRICHODERMA* CELLULASE**

PhD DISSERTATION

AYE AYE MYINT

**DEPARTMENT OF CHEMISTRY
UNIVERSITY OF YANGON
MYANMAR**

DECEMBER , 2004

Ph.D (Chemistry)**University of Yangon**

Title : Study on Production of Glucose from Cellulosic Wastes
Using Immobilized *Trichoderma* Cellulase

Candidate : Aye Aye Myint, M.Res (Chemistry)

Supervisors : Dr. Maung Maung Htay
Professor, Head,
Department of Chemistry, University of Yangon

Dr. Hnin Pwint Aung
Associate Professor,
Department of Chemistry,
University of Distance Education, Yangon

Dr. Saw Hla Myint
Associate Professor,
Department of Chemistry, University of Yangon

Dr. Myo Myint
Deputy Director,
Development Center for Pharmaceutical Technology,
Ministry of Industry (1), Yangon

Abstract : In the present work, cellulolytic fungi T_M (Myanmar soil) and T_Z (Azotobacter composte, China) were identified as *Trichoderma harzianum* by cultural, physiological and morphological characterization. The maximum specific activities of *Trichoderma harzianum* cellulase E_M (Myanmar) and E_Z (Azotobacter, China) were found to be $40.25 \times 10^{-2} \text{ Umg}^{-1}$ protein and $47.29 \times 10^{-2} \text{ Umg}^{-1}$ protein, respectively on the fifth day of fermentation at pH 6.5.

Five different cellulase samples: *Trichoderma viride* E_S (Sigma), E_B (Boehringer), E_T (Commercial), *Trichoderma harzianum* E_M (Myanmar) and E_Z (Azotobacter, composte) were used to detect cellulase activity quantitatively. More endo-glucanase activities of all enzymes on CMC (carboxymethyl cellulose) substrate than exo-glucanase activities on cotton and Avicel were observed.

Various types of five enzymes were immobilized by gel entrapment method using 4% sodium alginate solution and 0.2M calcium chloride

solution. The recommended optimal pH and temperature of free and immobilized samples were found to be the same at pH 5.5 and 50°C. The decreased K_m (Michaelis Menten const.) values were observed in all immobilized enzyme samples at optimal condition.

Saccharification of five free enzymes using cellulose powder, newspaper, packing paper, saw dust and rice husk were also employed in this research. The yield of glucose produced from newspaper at optimal condition by five enzyme samples were E_B (10.05%), E_S (8.46%), E_T (21.51%), E_M (3.14%) and E_Z (5.50%) whereas that of glucose produced from packing paper were E_B (12.97%), E_S (12.22%), E_T (28.55%), E_M (6.32%) and E_Z (6.02%) respectively. The yield of glucose produced from newspaper and packing paper were greater than saw dust and rice husk in case of all five enzymes subjected. For instance, the yield of glucose produced from newspaper (8.46%) and packing paper (12.22%) were greater than sawdust (4.45%) and rice husk (7.21%) when enzyme E_S (Sigma) was employed. The production of glucose by immobilized cellulase E_S (Sigma) at 27°C were found to be 3.74% from newspaper and 2.42% from packing paper which were less than the free form 7.24% and 10.22%.

The stability of immobilized cellulase E_M (Myanmar) showed 50% on the third cycle whereas E_S (Sigma) was still active 62% up to third cycle. The long-term stability of immobilized cellulase E_S (Sigma) kept for two weeks at 4°C was found to be active 1% while the activity of E_M (Myanmar) was found to be 2% after five days storage. This finding can be utilized in the large scale production of glucose from cellulosic wastes applying the improved method of cellulase immobilization.

Keywords : *Trichoderma*, cellulase, immobilized, saccharification, CMC