Cloning of β -glucosidase gene from Cellulomonas sp. into E.coli

To clone β -glucosidase gene from Cellulomonas sp. a gene library was constructed using E. coli JM83 pUC9. Among 2,500 pseudotransformants obtained, 20 clones developed yellow color on the p-nitrophenyl- -D-glucopyranoside filter paper. These 20 clones were classified into three groups based on the results of activity staining using nondenaturating polyacrylamide gel electrophoresis and restriction enzyme digestions. Among the three groups, only one group containing pCEl plasmid has specificity for cellobiose.

Purification and reaction pattern of cephalexin synthesizing enzyme from Acetobacter turbidans

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Cephalexin synthesizing enzyme (α amino acid ester hydrolase) was partially purified from the culture broth of *Acetobacter turbidans* ATCC9325 through ammonium sulfate fractionation, DEAE, CM, and Sephacryl S-200 gel filtration. The enzyme has optimum pH 6.0 and temperature, 40°C respectively. From the analysis of reaction mixtures by thin layer chromatographic and high performance liquid chromatographic techniques, it was confirmed this enzyme catalyzed simultaneously the following reactions:

- 1) Synthesis of cephalexin from D- α -phenylglycine methylester (PGM) and 7-amino 3-deacetoxy-cetoxycephalosporanic acid (7-ADCA)
- 2) Hydrolysis of cephalexin to form 7-ADCA and phenylglycine (PG)
- 3) Hydrolysis of PGM to form PG and methanol.

Base on the above experimental observations, the reaction model of this enzyme was identical with that of the enzyme from Xanthomonas citri.

Fermentation of carboxymethylcellulase using recombinant DNA-Bacillus megaterium

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For the analysis of fermentation characteristics and productivity of plasmid coded product, carboxymethylcellulase in a recombinant DNA cell fermentation system, batch and continuous fermentations were carried out using a *Bacillus* megaterium ATCC 14945 transformed with a plasmid, pCK 108 haboring carboxymethyl cellulase gene. The effects of carbon and nitrogen sources and of temperature and pH on cell growth, product yield, plasmid stability, specific plasmid contents of cell,