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CULTURE CONDITION OPTIMIZATION AND CHARACTERIZATION OF FUNGAL CELLULASE ENZYME

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Cellulases are among the industrially important hydrolytic enzymes and are of great significance in present day biotechnology. Recently there has been a resurgence in utilization of biomass for fuel production employing cellulases, hence forth in obtaining enzymes with novel activities in better yield. Fungi are important in cellulase production because of their ability to produce whole cellulase enzyme complex and secrete extracellular enzyme in high yield. This study was aimed at identification of a soil isolated fungal species which is able to produce cellulase enzyme and characterization of the enzyme for optimum activity.

In a previous study several fungal species that were able to utilize cellulose as the substrate were isolated from soil samples using serial dilution method. Their ability to degrade cellulose was demonstrated through their growth in agar medium with added cellulose powder as the sole carbon source. One of the species was identified as *Penicillium* sp based on colony characters and through microscopic observations of vegetative and reproductive structures. Cellulolytic activity was qualitatively displayed through the clearing zones obtained when the fungal inoculated plates were treated with Gram's iodine solution. The crude enzyme was obtained by inoculating the fungus to liquid cellulose powder medium (CPM). In quantitative assay, the filter-paper-hydrolyzing (FPase) activity of crude enzyme was assessed by Dinitro salicylic acid (DNS) method. One unit of the enzyme was expressed as the milligrams of reducing sugar liberated per milliliter of enzyme per minute. Culture conditions were optimized for cellulase production by the *Penicillium* sp in varying cellulose concentrations, pH and nitrogen sources. Crude enzyme was fractionated using Diethylaminoethyl (DEAE) Cellulose column chromatography and the fractionated cellulase enzyme was characterized for pH, pH stability, temperature and for activators and inhibitors.

In culture condition optimization, highest cellulase enzyme production by *Penicillium* sp was obtained in 0.6% cellulose powder media at pH 2 in the presence of yeast extract as the nitrogen source. The optimum pH value for the fractionated cellulase was 8.0 – 9.0 and the enzyme was optimally active at temperature of 40 °C. The activity of the fractionated cellulase was stimulated greatly by low concentrations (1 mM) of Co^{2+} while Ethylenediamine-tetraacetic acid (EDTA) was found to inhibit enzyme activity at same concentration. The optimum stability of fractionated cellulase was attained at pH 9.

The findings of this study indicate the potential of the use of cellulase enzyme isolated from *Penicillium sp* in industrial purposes.