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Efficacy of *Trichoderma* spp. in cellulase enzyme complex production

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SUMMARY

The present study was carried out to analyze the production of cellulase enzyme complex like endo-β 1,4 glucanases (C_x cellulase) and exo-β 1,4 glucanases(C_1 cellulase) at extracellular and intracellular level at different concentration of carbon and nitrogen sources by cellulolytic fungi *Trichoderma viride,T.koningii and T.harzianum.*A significant increase in extracellular endo-b 1,4 glucanase activity was recorded in *T. viride* in 2 % glucose as carbon source (48.63 Ul^{-1}) and 91.76 Ul^{-1} in 3 % urea as nitrogen source and maximum intracellular endoglucanase activity was registered by *T. viride* 33.60 Ul^{-1} in 3 % glucose as carbon source and 96.46 Ul^{-1} in 3 % urea as nitrogen source. *T. viride* showed maximum exoglucanase activity of 45.86Ul^{-1} at 3 % concentration of maltose as carbon source and 86.73 Ul^{-1} at 3 % of urea as nitrogen source at extracellular level. A significant increase in exoglucanase activity was registered by *T. viride* in 3 % glucose as carbon source (32.66Ul^{-1}) and 3 % urea as nitrogen source (119.86Ul^{-1}) at intracellular level when compared to the other fungal samples.

Key words : *Trichoderma* spp., Endoglucanase, Exoglucanase

Cellulose occurs as the structural element of plants and is thus present as a major component in agricultural and municipal waste and is the earth's most abundant renewable resource. Two main obstacles hindering the efficient transformation of cellulose are the highly ordered crystalline cellulose structure and a lignin seal usually surrounding cellulose fibers.Cellulose is commonly degraded by an enzyme called cellulase.The cellulase is a complex system comprised mainly of three enzymes endo-β-glucanase, exocellobiohydrolase and β-glucosidase. These enzymes comprise together a system to convert cellulose to glucose.Flilamentous fungi particularly *Aspergillus* spp. and *Trichoderma* spp. are efficient producers of cellulase enzyme complex,which degrades cellulose into soluble sugar glucose (Peij *et al*.,1998).Cellulase synthesis has been shown to be affected by various kinds of carbon and nitrogen sources , pH and surface active substances(Desai and Patel,1982).

Hence, the present investigation was carried out to analyze the efficacy of *Trichoderma* spp. (*T. viride,T. koningii and T. harzianum*)in the production of cellulase enzyme complex like endo-β 1,4 glucanases(C_x - cellulase) and exo-β 1,4 glucanases(C_1-cellulase) at extracellular and intracellular level at different concentration of carbon and nitrogen sources.

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MATERIALS AND METHODS

T.viride, T.koningii and T.harzianum were bought from Institute of Microbial Technology, Chandigarh, India. Fresh spent mushroom substrate samples were collected from Tamil Nadu Agricultural University, Coimbatore.

Growth medium for fungal culture:

Potato dextrose agar medium (PDA)- (Riker and Riker, 1936)*:*

From peeled potato (250 g), PDA medium was prepared by adding dextrose (20 g) and agar (15 g) .The volume was made into 1000 ml with distilled water and sterilized. The medium was poured into sterilized Petriplates (15 ml/ plate).Under aseptic conditions, *Trichoderma* spp. were inoculated. The bacterial growth was suppressed by the addition of 1ml of 10,000 ppm streptomycin sulphate solution and growth pattern was noticed.

Enzymology:

Preparation of culture medium:

Czapex-dox liquid medium(Raper and Thom, 1949)

Czapek-dox liquid medium was prepared by adding cellulose(10g), sodium nitrate(2.0 g),potassium chloride (0.5 g),magnesium sulphate (0.5 g), dipotassium hydrogen phosphate (1.0 g), ferrous sulphate (0.01 g) in 1000 ml distilled water with 1.0 ml of trace metal solution comprising of zinc sulphate and copper sulphate (1.0 g and 0.5 g dissolved in 100 ml distilled water).About fifty ml of Czpek- dox liquid medium was dispensed in 250ml Erlenmeyer flasks and sterilized at 1 atm for 15 minutes. After cooling, one ml of streptomycin sulphate (10,000 ppm) was added. The pH of the medium was maintained