Environment & Ecology 27 (2) : 689—691, 2009
 ISSN 0970-0420

Cellulolytic Enzyme Profile of Different Mushrooms Species on Maize Residue

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Abstract

The production of cellulases (carboxymethylcellulase, filterpaperase, cellobiase) and xylanase by different mushroom strains on the Reese mineral medium supplemented with maize residue as carbon source at 7 and 15 days of incubation was observed. The cultural filtrate of these mushroom strains exhibited relatively higher activity of all four enzymes at 15 days than that at 7 days of submerged fermentation.

Key words : Cellulases, Xylanase, Maize residue, Mushrooms.

Lignocellulose represents the most abundant renewable organic matter on the earth (1). Lignocellulose is main source of natural cellulose generated in agriculture, food processing and municipal service (2). Celluloytic enzymes play an important role in natural biodegradation process. Mushrooms are well known to secrete wide range of extra-cellular lignocellulolytic enzymes. These extra-cellular enzymes are attaching on the lignocellulosic material and convert complex organic matter into soluble substances, which can then be absorbed by the mushrooms for the purposes of their growth and development of fruiting bodies. For the conversion of cellulose to glucose, hydrolytic enzymes viz. endo-β-1,4 glucanase (EC 3.2.1.4), exo-β−1,4 glucanase (EC 3.2. 1.91) and β−1,4 glucosidase (EC 3.2.1.21) are involved. Cellulolytic enzymes have numerous application and biotechnological potential for various industries including chemicals, fuel, food, brewery, wine, animal feed, textile, laundry, pulp and paper and agriculture (1, 3). The present study was undertaken to assess the cellulolytic activity of eight different strains of mushrooms in submerged fermentation for 7 and 15 days of incubation on the Reese mineral medium supplemented with maize residue.

(Authors are thankful to Dean, Rajasthan College of Agriculture, for providing necessary laboratory facilities at Department of Molecular Biology and Biotechnology, RCA, Udaipur).

Methods

Pure cultures of eight different strains of mushroom were selected for this study. These were *Agaricus bisporus, Auricularia polytricha Ganoderma lucidium, Hypsizygus ulmarius, Lentinula edodes, Pleurotus florida, Pleurotus fossulatus* and *Pleurotus sajorcaju.* There were obtained from All India Co-ordinated Research Project on Mushroom , College of Agriculture, Udaipur, Rajasthan. Mycelial discs of 6 mm diameter from five days old culture of different fungi were inoculated in 250 ml sterilized Erlenmeyer flasks which contain 100 ml Reese mineral medium supplemented with maize residue (1% wt/vol.) as sole carbon source. After 7 and 15 days of incubation the culture filtrates were obtained and used for assay of extracellular enzyme activity. The carboxymethylcellulase, filter paperase and cellobiase activity were assayed by method of Ghose et al. (4). Xylanase was estimated by method of Reese and Mandel (5). The released reducing sugar was estimated by Miller's method (6). The enzyme activity of carboxymethylcellulase, filterpaperase, cellobiase, xylanase was expressed in IU/mg of total soluble protein content of the crude enzyme filtrate.

Results and Discussion

The results presented in Table 1 revealed that

Table 1. Enzymes activity (IU/mg) of mushroom strains after 7 days of growth on Reese's mineral medium supplemented with maize residue as sole carbon source.

Mushroom strains	Carboxy-methyl cellulase	Fil-terpa-perase	Cello-biase	Xyla-nase
Agaricus biaporus	0.210	0.094	0.041	0.175
Auricularia polytricha	0.096	0.044	0.029	0.133
Ganoderma lucidium	0.117	0.092	0.013	0.089
Hyzpizygus ulmarius	0.133	0.092	0.038	0.140
Lentinula edodes	0.124	0.047	0.032	0.219
Pleurotus florida	0.076	0.035	0.022	0.093
Pleurotus fossulatus	0.166	0.092	0.020	0.172
Pleurotus sajor-caju	0.113	0.077	0.021	0.133
CD at 5%	0.006	0.006	0.015	0.016
SE ±	0.002	0.001	0.005	0.005

Table 2. Enzymes activity (IU/mg) of mushroom strains after 15 days of growth on Reese's mineral medium supplemented with maize residue as sole carbon source.

Mushroom strains	Carboxy-methyl cellulase	Filter-pape-rase	Cello-biase	Xyla-nase
Agaricus biaporus	0.491	0.763	0.043	0.299
Auricularia polytricha	0.374	0.446	0.033	0.172
Ganoderma lucidium	0.275	0.392	0.019	0.130
Hyzpizygus ulmarius	0.498	0.356	0.038	0.278
Lentinula edodes	0.913	0.364	0.026	0.328
Pleurotus florida	0.156	0.329	0.025	0.153
Pleurotus fossulatus	0.535	0.370	0.027	0.305
Pleurotus sajor-caju	0.417	0.589	0.021	0.200
CD at 5%	0.009	0.041	0.014	0.019
SE ±	0.003	0.012	0.003	0.008

after 7 days of incubation in maize residue maximum carboxymethylcellulase activity was found in *Agaricus bisporus* (0.210 IU/mg) followed by *Pleurotus fossulatus* (0.166 IU/mg). The maximum filter paperase activity was recorded in *A. bisporus* (0.094 IU/mg) followed by *H. ulmarius* and *Pleurotus fossulatus* (0.092 IU/mg). The minimum carboxymethylcellulase and filter paperase activity were recorded with *P. florida* (0.076 and 0.35 IU/mg respectively). The maximum cellobiase activity was found in *A. bisporus* (0.041 IU/mg) followed by *H. ulmarius* (0.038 IU/mg). The maximum activity of xylanase enzyme was assayed in *Lentinula edodes* (0.219 IU/mg) followed by *A. bisporus* (0.175 IU/mg). The minimum activity of cellobiase and xylanase was observed in *G. lucidium* (0.013 and 0.089 IU/mg respectively).

Cellulolytic and xylanolytic enzyme activity of all the mushrooms strains increased after 15 days of incubation in Reese's mineral medium supplemented with maize residue as shown in Table 2. Maximum carboxymethylcellulase activity was assayed in *Lentinula edodes* (0.913 IU/mg) followed by *Pleurotus fossulatus* (0.535 IU/mg). The maximum filter paperase activity was recorded in *A. bisporus* (0.763 IU/mg) followed by *Pleurotus sajor-caju* (0.589 IU/mg). The cellobiase activity was found to be maximum in *A. bisporus* (0.043 IU/mg). Xylanase activity was highest in *L. edodes* (0.328 IU/mg). Even after 15 days of incubation, the minimum carboxymethylcellulase and filterpaperase enzyme activity were assayed in *P. florida* while minimum activity of cellobiase (0.019 IU/mg) and xylanase (0.130 IU/mg) was assayed in *G. lucidium.*

Lower activity of cellulolytic and xylanolytic enzymes of *P. florida* and *G. lucidium* at both 7 and 15 days of incubation may be responsible for their slow growth on lignocellulosic materials (Tables 1 and 2).

Similar observations on cellulolytic enzyme activity of different mushrooms and other fungi were reported by many workers. Vijaya and Singaacharya (7) reported the cellulolytic activity of *Pleurotus ostreatus* as carboxymethylcellulase (28 relative enzyme activities) and filterpaperase (336 mg/ml) on the paddy straw. Singh et al. (8) reported extra-cellular activity in P- 9 isolated of *Morchella esculanta*; 0.29 to 0.554/ml of endo-glucanase, 0.61 to 0.77 U/ml of β-glucosidase and 0.71 to 0.94 U/ml of xylanase on different combination of wheat straw and wheat flour. In *Phanerocnaete chrysorium* maximum FPU/ml was reported to be 2.04/ml on untreated rice husk, 1.92 U/ml on untreated wheat straw at 8 days of incubation (9). Madhurendra et al. (10) reported the cellulase activity of *Pleurotus* sp. on barley based straw in which exo-β-1, 4 glucanase activity was found to be 6 unit/mg protein by *P. florida* and 4 unit/mg of protein by *P. sajor-caju* at 6 days of incubation on barley based straw. After 15 days of incubation the exo-β–1, 4 glucanase was reported to be 6 unit/mg of protein by *P. florida* and 6 unit/mg of protein by *P. sajor-caju*. The endo-β-1,4 glucanase activity was found to be 17 unit/mg protein by *P. florida* and *P.sajor caju* at 6 days of incubation. At 15 days of incubation 39 unit/

mg protein by *P. florida* and 44 unit/mg protein by *P. sajor-caju* were recorded. The variations observed in present investigation may be attributed to specific strains and the substrate used for the submerged growth of mushrooms. Production of lignocellulolytic enzymes by different fungi may vary because of the influence of substrate (carbon source) and atmospheric conditions on the growth of organism.

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