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Applied Botany

Elixir Appl. Botany 34 (2011) 2543-2544

ISSN: 2229-712X

Cellulase enzyme complex and xylanase enzyme profile in the basidiocarp of mushrooms

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ARTICLE INFO

Article history:

Received: 16 March 2011;

Received in revised form:

23 April 2011;

Accepted: 28 April 2011;

Keywords

Exo β-1,

4 glucanase,

endo β-1,

4 glucanase,

β-glucosidase and

xylanase.

ABSTRACT

A laboratory investigation was undertaken to explore the production of cellulase enzyme complex and xylanase enzyme in the Boletus edulis, Ganodrema tsugae and Micoporus xanthopus. The result of the study revealed that the exo-B 1,4 glucanase activity, endo B-1,4 glucanase basidiocarp of mushrooms, viz., activity, B-glucosidase activity and xylanase activity were very much pronounced in Ganoderma tsugae (1.791 Umg-1 1.864 Umg-1,1.127 IUmg-1 and 0.142 IUmg-1 enzyme protien) than Boletus edulis (0.555 Umg-1,1.05 Umg-1,0.683 IUmg-1 and 0.063 IUmg-1 enzyme protein)and Microporus xanthopus (1.142Umg-1,1.503 Umg-1, 0.623 IUmg-1& 0.038 IUmg-1 enzyme protein).

Introduction

White-rot basidiomycetes are efficient decomposers of lignocellulose, due to their capability to synthesize relevant hydrolytic and oxidative extracellular enzymes. Lignocellulolytic enzymes have significant potential applications not only in the chemical, fuel, food, textile, laundry and pulp and paper industries but also in agriculture and for animal feed production (Elisashvili *et al*., 2006).

Ganoderma species, basidiomycetous fungi belongs to the order Polyporales (Chang, 1995 and Wasser and Weis, 1999). *Ganoderma tsugae* is recognized by its varnished, reddish cap and stem. The cap is 5 – 30 cm at first, irregularly knobby or elongated, but by maturity more or less fan-shaped with a shiny, varnished surface often roughly arranged into lumpy "zones". The cap is red to reddish brown when mature and when young often with zones of bright yellow and white toward the margin and occasionally with bluish tints. The stem is 3 – 14 cm long, upto 3 cm thick, twisted, equal or irregular, varnished and coloured like the cap and often distinctively angled away from one side of the cap.

Microporus xanthopus also belongs to the order polyporales is found on rotting wood. The mature fruiting bodies have thin, funnel shaped caps that are concentrically zoned in various shades of brown and are supported by a yellow footed stem. On the white underside of the pileus, there are numerous tiny pores. The fruiting body appears as a white fleck on the wood surface. This enlarges into a hemispherical cushion between half a millimetre and a millimetre wide. Then the primordium elongates via vigorous growth to develop the stem. Eventually the hyphae in the central part of the stem apex stop growing while those on the periphery continue to grow and so develop the funnel-shaped pileus.

Boletus edulis, commonly known as *porcini* or *cep*, a basidiomycete fungus belongs to the order Boletales. The fruit body consists of a large and imposing brown cap which on occasion can reach at least 35 cm in diameter and 3 kg in weight. Like other *Boletus*, it has tubes extending downward from the underside of the cap, rather than gills. The stout stipe, is white or yellowish in colour, upto 25 cm tall and 10 cm thick, and partially covered with a raised network pattern or reticulations.

The aim of present investigation was to estimate the production of cellulase enzyme complex consisting of exo, endo–β–1, 4 glucanase and β–glucosidase (filter paper activity) and xylanase enzyme activity.

Materials and methods

Collection and preparation of samples

The basidiocarp of *Boletus edulis*, *Ganoderma tsugae* and *Microporus xanthopus* were collected from wooden logs in Kannur district, Kerala State and were identified based on the morphology of the pileus and stipe of the basidiocarp.The basidiocarp of the mushroom sample was shade dried and powdered. The powdered sample was used for the study.

Preparation of enzyme assay from fungal basidiocarp(*boletus edulis, ganoderma tsugae* and *microporus xanthopus)*

The basidiocarp of the fungus was washed twice with distilled water. A quantity of 1.0 g of powdered basidiocarp was macerated in five ml of sodium acetate-acetic acid buffer (pH 5.2) for endoglucanase assay, five ml of sodium citrate buffer (pH 5.0) for exoglucanase assay, 5 ml of sodium acetate-acetic acid buffer(pH 5.8) for β-glucosidase in a pre-chilled porcelain mortar and pestle with a pinch of acid washed sand. The homogenate was centrifuged in a refrigerated centrifuge at 10,000 x g for 15 minutes.

The supernatant served as enzyme source. Enzyme extraction, processing and other procedures were done at 5°C in a cold room.

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Results and discussion

Cellulase enzyme complex (table-1)

Among the cellulase enzyme complex, endo-β-1, 4 glucanase activity in the basidiocarp powder of *G. tsugae* was found to be more pronounced (1.864 U mg^{-1} enzyme protein) than *B. edulis* (1.051 U mg^{-1} enzyme protein). The exo-β-1, 4 glucanase activity was expressed maximally, 1.791 U mg^{-1} enzyme protein in *G. tsugae* and minimally in *B. edulis*, (0.555 U mg^{-1} enzyme protein). The basidiocarp of *G. tsugae* registered a highest β-glucosidase enzyme activity of 1.127 IU mg^{-1} enzyme protein in comparison to *M. xanthopus*, 0.623 IU mg^{-1} enzyme protein.

Similar trend of result was reported by Kamal *et al*. (2000) with the endoglucanase activity of 0.085 U ml^{-1} and β-Glucosidase activity of 1.008 U ml^{-1} in *Agaricus bisporus*. In *Volvariella volvacea*, Choudhary *et al*. (2009) and in *Pleurotus florida*, Mishra (2009) also obtained more or less same level of activity of endoglucanase and cellobiase enzymes.

Similar finding was recorded by Choudhary *et al*. (2009). They obtained endoglucanase, filter paper and cellobiase enzyme activities of 1.952, 3.36 and 4.18 IU / 100 ml respectively in *Volvariella volvacea* (paddy straw mushroom).

Mishra (2009) also reported the production of 0.144 U ml^{-1} of endoglucanase, 0.137 IU ml^{-1} filter paper activity and 0.041 IU ml^{-1} of cellobiase enzyme activities in *Pleurotus florida* after 15 days of incubation with carboxymethyl cellulose as carbon source.

Xylanase enzyme (table-1)

The highest xylanase enzyme activity of 0.142 IU mg^{-1} enzyme protein was registered by *Ganoderma tsugae* basidiocarp and the least activity of 0.038 IU mg^{-1} enzyme protein was recorded by *Microporus xanthopus*.

The present result is in agreement with the result of Vijaya and Singaracharya (2005). Who have obtained 0.094 IU ml^{-1} of xylanase enzyme activity in *Pleurotus ostreatus* on different combinations of wheat straw and wheat flour.

Similar finding was reported by Choudhary *et al*. (2009). They have obtained xylanase enzyme activity of 0.57 IU ml^{-1} during the biodegradation of paddy straw by *Volvariella volvacea*.

Conclusion

The mushroom described as "precious pearls of cookery" can be effectively harnessed for maximum production of cellulolytic enzymes. The development of stable, highly active microbial oxidative enzymes have long been an elusive goal of biotechnology for a wide variety of commercially attractive processes, from polymer synthesis to bioremediation.

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Table – I Cellulase Enzyme Complex And Xylanase Enzyme Activity

S.No.	Mushroom	Cellulase enzyme complex			Xylanase**
		Endo-β-1*, 4 glucanase	Exo-β-1*, 4 glucanase	β-glucosidase**	
1.	*Ganoderma tsugae*	1.864	1.791	1.127	0.142
2.	*Microporus xanthopus*	1.503	1.142	0.623	0.038
3.	*Boletus edulis*	1.051	0.555	0.683	0.063
SED		0.010	0.008	0.007	0.004
CD (P < 0.01)		0.036	0.028	0.027	0.016

* Enzyme activity expressed in U mg^{-1} enzyme protein.
** Enzyme activity expressed in IU mg^{-1} enzyme protein