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Short Note

Effect of medicinal plant extracts on mycelial growth of *Pleurotus florida*

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Mushrooms are considered as a delicacy with a high nutritional and functional value and accepted as functional and nutraceutical food ingredients. *Pleurotus* is a widely cultivated edible mushroom on a variety of substrates and conditions which have an impact on the content of macronutrients and mycochemicals. Nowadays, mushroom extracts are commercialized as dietary supplements, mainly for their anti-tumour activity and the enhancement of immune function. There has been an increasing interest in the use of *Pleurotus* spp. to supplement processed foods, such as bread and dairy products due to its potential to enhance protein content and quality, along with the beneficial health effects of their mycochemicals (Moon and Lo, 2014). Medicinal plants are frequently used for the isolation of biologically active compounds widely used in the preparation of various drugs. The use of substrates containing bioactive compounds or supplemented with minerals, as well as postharvest treatments are some of the strategies to increase the nutraceutical value of *Pleurotus* spp. (Carrasco-Gonzalez *et al*., 2017). The enrichment with medicinal plants *viz*., lemon grass, neem, citrus lemon and eucalyptus @ 4 per cent in substrate compost for the cultivation of *Pleurotus ostreatus* and *P. florida* revealed that the addition not only enhanced the yield but also reduced the incidence of contaminants. The addition of medicinal plants to substrate was preferable than chemicals due to enhancement of bioactive compounds and reduction in the lethal effects during consumption of mushroom (Inam-Ul Haq *et al*., 2014). *Azadiracta indica* (3%) amended beds showed maximum increase in yield of *Pleurotus* spp. and minimum disease incidence when compared to control (Shah *et al*., 2011). Owaid *et al*. (2019) observed that addition of 20 per cent leaf extracts of *Ficus caricae* and *Olea europiae* enhanced the mycelial growth of *P. ostreatus, P. cornucopiae, Coriolus versicolor and Ganoderma lucidum* under *in vitro* condition.

Collection and preparation of medicinal plant extracts

Medicinal plants *viz*., Adathoda (*Justicia adathoda*), Neem (*Azadiracta indica*) and Ocimum (*Ocimum sanctum*) were collected, washed, dried and powdered for amending the media (PDA). The medicinal plant extract was prepared by maceration method. The required concentrations of 250, 500 and 1000 ppm were prepared from the dried powder of the three medicinal plants in 50 ml sterile water respectively and kept in rotary shaker for 30 minutes. The suspension was filtered through Whatman No.1 filter paper followed by syringe filtration. The final suspension was used for further studies.

In vitro evaluation of the medicinal plants on mycelial growth of *P. florida*

The *in vitro* evaluation was carried out by poisoned food technique (Nene and Thapliyal, 1993). Double strength potato dextrose agar (PDA) was prepared @ 50 ml per conical flask and sterilized. To the molten medium on cooling, 50 ml of extract of respective medicinal plant extract (250, 500 and 1000 ppm solution of Adathoda, Neem and Ocimum) was added. The amended medium was mixed thoroughly and poured into sterile Petri dishes. Five mm discs

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Table 1. Nature of mycelial growth of *Pleurotus florida* in amended PDA medium

Treatments	Nature of mycelial growth		
	3 DAI	5 DAI	7 DAI
T1 (Adathoda at 250 ppm)	White sparse growth with fluffy centre and regular margin	White sparse growth with regular margin	White fluffy growth with regular margin
T2 (Adathoda at 500 ppm)	White growth with fluffy centre and regular margin	White sparse growth with regular margin	White fluffy growth with regular margin
T3 (Adathoda at 1000 ppm)	White growth with fluffy centre and regular margin	White fluffy growth with regular margin	White fluffy growth with regular margin
T4 (Neem at 250 ppm)	White cottony growth with fluffy centre and regular margin	White fluffy growth with regular margin	White fluffy growth with regular margin
T5 (Neem at 500 ppm)	White cottony growth with fluffy centre and regular margin	White fluffy growth with regular margin	White fluffy growth with regular margin
T6 (Neem at 1000 ppm)	White cottony growth with fluffy centre and regular margin	White fluffy growth with regular margin	White fluffy growth with regular margin
T7 (Ocimum at 250 ppm)	White growth with fluffy centre and regular margin	White fluffy with radiating growth and regular margin	White fluffy growth with regular margin
T8 (Ocimum at 500 ppm)	White growth with fluffy centre and regular margin	White fluffy with radiating growth and regular margin	White fluffy growth with regular margin
T9 (Ocimum at 1000 ppm)	White growth with fluffy centre and regular margin	White fluffy with radiating growth and regular margin	White fluffy growth with regular margin
T10 (Control)	White sparse growth	White sparse mycelial growth	White fluffy growth with regular margin

Table 2. Growth of mycelium of *P. florida* in medicinal plant amended PDA medium (3, 5 and 7 DAI)

Treatments	Radial growth of mycelium in Petri dish (DAI)*			Rate of growth	DTCP**
	3rd	5th	7th	(cm day $^{-1}$)	
T1	2.60 (1.61) ^{bcde}	5.73 (2.39) ^{abc}	8.88 (2.98) ^{ab}	1.57	7
T2	2.70 (1.64) ^{ab}	5.78 (2.40) ^{a}	8.88 (2.98) ^{ab}	1.55	7
T3	2.75 (1.66) ^{a}	5.85 (2.42) ^{a}	8.90 (2.98) ^{ab}	1.57	7
T4	2.40 (1.55) ^{f}	5.43 (2.32) ^{d}	8.40 (2.90) ^{c}	1.53	8
T5	2.65 (1.63) ^{abc}	5.45 (2.33) ^{d}	8.53 (2.92) ^{c}	1.47	7
T6	2.75 (1.66) ^{a}	5.75 (2.34) ^{ab}	8.80 (2.96) ^{ab}	1.51	7
T7	2.48 (1.57) ^{ef}	5.80 (2.40) ^{a}	8.83 (2.97) ^{ab}	1.58	7
T8	2.50 (1.58) ^{def}	5.68 (2.37) ^{bc}	8.78 (2.96) ^{b}	1.56	7
T9	2.52 (1.59) ^{cdef}	5.58 (2.36) ^{cd}	8.75 (2.95) ^{b}	1.55	7
T10(Tc)	2.63 (1.62) ^{abcd}	5.90 (2.45) ^{a}	9.00 (3.00) ^{a}	1.60	7
SEm±	0.038	0.049	0.054	0.047	
CD (0.05)	0.132	0.166	0.202		

* Mean of four replications; **Days taken for completion of growth in petri dish;
Values in parenthesis are square root transformed values; DAI- Days after inoculation

were cut from the periphery of actively growing *P. florida* culture with a sterile cork borer and laid aseptically at the centre of each Petri dish. The experiment was laid out in Completely Randomized Design with ten treatments (control + 9 treatments representing three medicinal plants each at three doses) and four replications. The inoculated dishes were incubated under room temperature (25±2° C) and observed for nature of mycelial growth (colour and pattern), rate of mycelial growth and days taken for complete mycelial growth. The mycelial growth rate was also assessed. The data obtained were subjected to analysis of variance (ANOVA) and critical difference (CD) was calculated at five percent level of significance for comparison among different treatments.

The results of the *in vitro* evaluation revealed that the mycelium of *P. florida* in the amended media showed variations in pattern and density. In adathoda amended medium at different concentrations, initially it produced a white sparse growth with regular margin turning to white fluffy growth with regular margin on completion of growth. For neem amended media, it produced a white cottony growth with regular margin three days after inoculation eventually turning to white fluffy growth with regular margin. In ocimum amended media, the growth was white sparse with fluffy centre and regular margin later turning to white fluffy with regular margin. In control, initially white sparse growth was noticed which later turned white cottony growth with regular margin. The radial growth of mycelium indicated that the growth of *P. florida* in amended medium was at par with control (Table 1, Fig. 1).

The amendments at different concentrations were not inhibitory for the mycelial growth of *P.florida*. The radial growth of mycelium in Petri dish after 3, 5 and 7 days after inoculation and statistical differences in growth on different days are presented in Table 2. The amendments *viz*., adathoda, neem and ocimum showed enhanced mycelial growth with increase in concentration. The radial growth of mycelium 5 DAI in the various treatment ranged from 5.43-5.90 cm with maximum growth in control. Seven days after inoculation all the treatments were at par with control which recorded a maximum growth of (9.0 cm) (Table 2). Kumar *et al.* (2019) evaluated the effect of leaf extracts *viz.,* Neem, Eucalyptus and Lantana at 2 and 4 per cent concentrations on the mycelial growth of *Pleurotus sapidus*. Leaf extract of lantana at 4 per

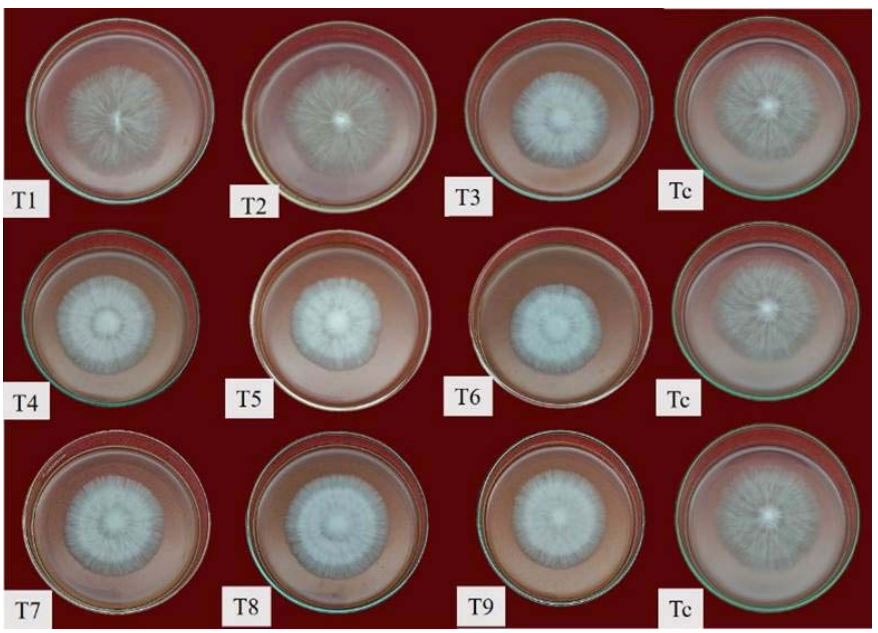

Fig. 1. Mycelial growth of *P. florida* in amended PDA medium (5 DAI)

cent and 2 per cent gave a higher radial mycelial growth. The leaf extract of Eucalyptus inhibited the mycelial growth at both 2 and 4 per cent. Pandey *et al*. (2021) also observed increase in mycelial growth of *P. florida* using neem leaf extract while eucalyptus had an inhibitory effect on growth of the mycelium. The rate of growth in different treatments ranged from 1.47- 1.60 cm day $^{-1}$. The days taken for complete growth in Petri dish was 7 days except for neem at 250 ppm, which took 8 days. In the present study it was concluded that selected medicinal plants didn't have inhibitory effect on the mycelial growth of *P. florida* under *in vitro* condition and thus can be studied for development of enriched mushrooms. Biomedicinal enrichment of substrate may be a stepping-stone in the development of mushroom nutraceuticals.

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