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# Cultivation of *Pleurotus ostreatus* on weed plants

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**Abstract**

Oyster mushroom, *Pleurotus ostreatus* (Jacq.:Fr.) Kumm. ITCC 3308 (collected from Indian Type Culture Collection, IARI, New Delhi, India, 110012) was grown on dry weed plants, *Leonotis* sp, *Sida acuta*, *Parthenium argentatum*, *Ageratum conyzoides*, *Cassia sophera*, *Tephrosia purpurea* and *Lantana camara*. *Leonotis* sp. was the best substrate in fruit body production of *P. ostreatus* when it was mixed with rice straw (1:1, wet wt/wet wt) for mushroom cultivation. The fruiting time for *P. ostreatus* was also less on *Leonotis* sp. than on any other weed substrates tested in the present investigation. *T. purpurea* was the least suited weed for oyster mushroom cultivation. The main problem of oyster mushroom cultivation on weed substrates was found to be low yield in the second flush that could be overcome by blending weed plants with rice straw. The protein contents of the fruit bodies obtained from *Cassia sophera, Parthenium argentatum* and *Leonotis* sp. were not only better than rice straw but also from the rice straw supplemented weeds.


*Keywords:* Cultivation; Mushroom; *Pleurotus ostreatus*; Rice straw; Weed

## 1. Introduction

Oyster mushroom (*Pleurotus* spp.) cultivation has increased tremendously throughout the world during the last few decades (Chang, 1999; Royse, 2002). This mushroom accounted for 14.2% of the total world production of edible mushroom in 1997 (Chang, 1999). Although commonly grown on pasteurized straw of wheat or rice, they can be cultivated on wide variety of substrates that contain lignin and cellulose. Oyster mushroom cultivation can play an important role in managing organic wastes which have become problematic for disposal.

The word 'weed' is generally used for the undesirable plants. Suitable utilization of weeds is a subject of interest as most weeds are not used even as fodder due to the presence of lignin and anti-metabolites like phenolics, glycosides, flavonoides and other compounds (Fianu et al., 1981). Disposal of these plants through burning causes environmental pollution as they release high level of $CO_2$ as well as it is the cause of unnecessary wasting of large amount of organic materials (Croan, 2000).

Oyster mushrooms (*Pleurotus* spp.) can produce fruit bodies on straws of rice (*Oryza sativa*), wheat (*Triticum vulgare*), ragi (*Elucine coracana*), bazra (*Pennisetum typhoides*), sorghum (*Sorghum vulgare*), maize (*Zea mays*) (Bano et al., 1987; Goswami et al., 1987), woods of poplar (*Populus robusta*), oak (*Quercus leucotrichopora*), horse chest nut (*Aesculus indica*), *Acasia* sp. (Pant et al., 1987), chopped banana pseudostem (Singh and Tandon, 1987), cotton stalk, pea shells and poplar saw dust (Philippoussis et al., 2001; Zervakis et al., 2001). In this paper we report that weeds can be used as the substrate for cultivation of oyster mushroom (*Pleurotus ostreatus*).

## 2. Methods

### 2.1. Mushroom strain

*Pleurotus ostreatus* (Jacq.:Fr.) Kumm. (ITCC 3308) was collected from Indian Type Culture Collection, Division of

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Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi, 110012, India and was maintained on potato-dextrose agar (pH 7.0) containing 20% potato extract; 2% dextrose; 2% agar as reported earlier (Das and Mukherjee, 1996).

### 2.2. Substrate preparations

Weed plants were collected after seed formation from local fields of Durgapur, West Bengal, India. The plants were *Leonotis* sp. (Lamiaceae), *Sida acuta* (Malvaceae), *Parthenium argentatum* (Asteraceae), *Ageratum conyzoides* (Asteraceae), *Cassia sophera* (Caesalpiniaceae), *Tephrosia purpurea* (Papilionaceae) and *Lantana camara* (Verbenaceae). The plants were completely dried in the sun after harvesting. Rice straw (var. MINIKIT) was obtained from a local farm at Durgapur, West Bengal, India and it was approximately three months in storage after harvesting. The branches of weeds without leaves or rice straw were chopped into small pieces (1–2 cm), weighed and soaked in water for overnight. Extra water present in the substrates was drained off and the substrates were spread on the surface of a clean blotting paper and air dried for 15 min to remove the excess water. No heat treatment of substrates was done. Wet (600 g) substrate (~85% moisture), which was either individual weed or combination of weed and rice straw (1:1) was mixed with 20% spawn (wet wt/ wet wt). The spawned substrates were then put into 30 cm × 42 cm polythene bags. The bags were tightly closed and pin holes were made on the surfaces. The bags were subsequently kept in a spawn running room at 25 ± 1 °C under dark condition until primordia was formed. After primordia formation, large holes were made in the polythene bags to allow the normal development of fruit bodies. The bags were then kept at 22 ± 1 °C with a 12 h photoperiod (1500–2000 Lux) and 85–90% relative humidity. Adequate ventilation was provided to prevent increase of $CO_2$ concentration. Mushrooms were harvested in clusters from the substrates manually, three days after primordia initiation. Biological efficiencies (BE) of mushrooms were calculated from the ratios of weight (kg) of fresh mushrooms harvested per kg of dry substrates.

### 2.3. Preparation of fruit body extract and measurement of protein

Fresh fruit bodies (28 g) after third day of primordia formation were disrupted by being crushed with acid-washed sand in mortar and pestle. The tissue was then extracted with 100 ml 20 mM imidazole buffer containing 1 mM EDTA, 2 mM 2-mercaptoethanol, 2 mM PMSF (pH 7.8). Unbroken cells and cell debris were removed by centrifugation at 32,000*g* for 30 min and the supernatant was used for analysis of protein. Protein concentrations were determined by the Bradford method (Bradford, 1976).

### 2.4. Statistical analysis of experimental data

The data obtained were analyzed using SPSS 13.0 version Software. Mean and standard deviations of 15 replications (3 sets × 5 batches) were calculated in each of the substrates. The means were sorted in decreasing order to show the absolute ranking of biological efficiencies of different substrates. 'Student's *t*-test' was performed at 5% level to check the level of significance of the differences between two consecutive substrates. A "statistical rank" data column has been provided to indicate whether there is statistically significant differences between the results in each set of substrates. Data followed by the same letter in statistical rank column indicates that there is no significant difference at 5% level according to 'Student's *t*-test'. All experiments were carried out using 15 replicates for each substrate.

## 3. Results and discussion

### 3.1. Fruiting efficiency of P. ostreatus on weeds

Biological efficiency (BE) of mushroom production and time for primordia initiation on different substrates are given in Tables 1 and 2. In the first flush of production, biological efficiency was better on *Leonotis* sp. than that with rice straw. However, the difference was apparent, not statistically significant as both the substrates belong to same statistical rank (Table 1). Biological efficiency increased significantly ($P < 0.05$) in comparison to rice straw or *Leonotis* sp. when both the substrates were mixed in 1:1 proportion for mushroom cultivation. Biological efficiency of the mushroom was found to be highest on combination substrates of rice straw and *Leonotis* sp. than on any weed or blended substrates not only in individual flush but also in accumulated conditions (Tables 1 and 2). Among the seven weeds tested for mushroom growth, *T. purpurea* was found to be the least responsive in terms of biological efficiency (Tables 1 and 2). However, addition of rice straw to *T. purpurea* showed an ameliorating effect on fruit body production when used as mushroom substrate. Here the biological efficiency of combination substrate uplifted to statistical rank 'C' from 'J' of the individual *T. purpurea* (Table 2). Other weeds showed a differential response but the trend of response in terms of biological efficiency was obvious when those were combined with rice straw (Table 2). In second flush the combination of weed and rice straw blends showed higher biological efficiency than any of the individual weed (Table 1). All the combined substrates in second flush belong to same statistical rank along with rice straw indicate that higher biological efficiency was due to the blending of rice straw with weeds. The increase of yields in the second flush might be due to higher water retention capacity of the combined substrates (particularly for rice straw) than that of individual weed substrate (data not shown). Among the different substrates primordia was formed most rapidly (9.66 days) either on *Leonotis* sp. or

Table 1
Biological efficiency of the cultivation of *Pleurotus ostreatus* mushroom (fresh mushroom in kg/kg dry substrate) on non-heat treated rice straw and weed species

| Biological efficiency in first flush (kg/kg substrate) | | | | | Biological efficiency in second flush (kg/kg substrate) | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Substrate | Mean[a] | Std. Deviation | *p*-value | Statistical rank | Substrate | Mean[a] | Std. Deviation | *p*-value | Statistical rank |
| Rice straw + *Leonotis* sp (1:1) | 0.943 | 0.0250 | | A | Rice straw + *Leonotis* sp. (1:1) | 0.447 | 0.0226 | | A |
| *Leonotis* sp. | 0.802 | 0.0349 | <0.05 | B | Rice straw | 0.437 | 0.0226 | >0.05 | A |
| Rice straw | 0.771 | 0.0405 | >0.05 | B | Rice straw + *Cassia sophera* (1:1) | 0.435 | 0.0119 | >0.05 | A |
| Rice straw + *Sida acuta* (1:1) | 0.749 | 0.0240 | >0.05 | B | Rice straw + *Tephrosia purpurea* (1:1) | 0.431 | 0.0329 | >0.05 | A |
| Rice straw + *Cassia sophera* (1:1) | 0.737 | 0.0324 | >0.05 | B | Rice straw + *Sida acuta* (1:1) | 0.421 | 0.0181 | >0.05 | A |
| *Ageratum conyzoides* | 0.696 | 0.0159 | <0.05 | C | Rice straw + *Ageratum conyzoides* (1:1) | 0.411 | 0.0146 | >0.05 | A |
| *Sida acuta* | 0.687 | 0.0247 | >0.05 | C | Rice straw + *Lantana camara* (1:1) | 0.411 | 0.0128 | >0.05 | A |
| Rice straw + *Ageratum conyzoides* (1:1) | 0.649 | 0.0285 | <0.05 | D | Rice straw + *Parthenium argentatum* (1:1) | 0.405 | 0.0125 | >0.05 | A |
| Rice straw + *Lantana camara* (1:1) | 0.647 | 0.0229 | >0.05 | D | *Leonotis* sp. | 0.222 | 0.0231 | <0.05 | B |
| Rice straw + *Tephrosia purpurea* (1:1) | 0.642 | 0.0224 | >0.05 | D | *Sida acuta* | 0.219 | 0.0133 | >0.05 | B |
| *Lantana camara* | 0.638 | 0.0218 | >0.05 | D | *Ageratum conyzoides* | 0.189 | 0.0110 | <0.05 | C |
| Rice straw + *Parthenium argentatum* (1:1) | 0.605 | 0.0196 | <0.05 | E | *Cassia sophera* | 0.183 | 0.0171 | >0.05 | C |
| *Cassia sophera* | 0.515 | 0.0229 | <0.05 | F | *Lantana camara* | 0.168 | 0.0101 | <0.05 | D |
| *Parthenium argentatum* | 0.480 | 0.0248 | <0.05 | G | *Parthenium argentatum* | 0.113 | 0.0111 | <0.05 | E |
| *Tephrosia purpurea* | 0.229 | 0.0299 | <0.05 | H | *Tephrosia purpurea* | 0.000 | 0.0000 | <0.05 | F |

[a] Means of 15 replications of each substrate were ranked according to Student's *t*-test at 5% level.

Table 2
Biological efficiency of the cultivation of *Pleurotus ostreatus* mushroom (fresh mushroom in kg/kg dry substrate) on non-heat treated rice straw and weed species

| Substrate | Biological efficiency of mushroom accumulating 1st and 2nd flush (kg/kg substrate) | | | | Time to primordia initiation | |
|---|---|---|---|---|---|---|
| | Mean[a] | Std. Deviation | *p*-value | Statistical rank | 1st Flush | 2nd Flush |
| Rice straw + *Leonotis* sp. (1:1) | 1.390 | 0.0307 | | A | 9.66 | 16.66 |
| Rice straw | 1.208 | 0.0544 | <0.05 | B | 15.33 | 22.33 |
| Rice straw + *Cassia sophera* (1:1) | 1.171 | 0.0374 | >0.05 | B | 17.66 | 25.66 |
| Rice straw + *Sida acuta* (1:1) | 1.170 | 0.0299 | >0.05 | B | 10.00 | 19.00 |
| Rice straw + *Tephrosia purpurea* (1:1) | 1.073 | 0.0213 | <0.05 | C | 13.66 | 21.66 |
| Rice straw + *Ageratum conyzoides* (1:1) | 1.060 | 0.0378 | >0.05 | C | 11.66 | 18.33 |
| Rice straw + *Lantana camara* (1:1) | 1.057 | 0.0324 | >0.05 | C | 13.66 | 20.66 |
| *Leonotis* sp. | 1.024 | 0.0485 | <0.05 | D | 9.66 | 17.66 |
| Rice straw + *Parthenium argentatum* (1:1) | 1.01 | 0.0275 | >0.05 | D | 15.00 | 22.00 |
| *Sida acuta* | 0.906 | 0.0287 | <0.05 | E | 10.00 | 18.00 |
| *Ageratum conyzoides* | 0.885 | 0.0223 | <0.05 | F | 11.33 | 18.33 |
| *Lantana camara* | 0.806 | 0.0256 | <0.05 | G | 15.66 | 22.66 |
| *Cassia sophera* | 0.696 | 0.0294 | <0.05 | H | 18.66 | 24.33 |
| *Parthenium argentatum* | 0.593 | 0.0304 | <0.05 | I | 16.66 | 24.66 |
| *Tephrosia purpurea* | 0.229 | 0.0299 | <0.05 | J | 14.33 | – |

– No fruit body was detected.
[a] Means of 15 replications of each substrate were ranked according to Student's *t*-test at 5% level.

*Leonotis* sp. supplemented with rice straw substrate in the first flush. Primordia formations on those two substrates were also found to take less time in the second flush as compared to other substrates used for mushroom cultivation (Table 2).

### 3.2. Protein concentrations in fruit bodies of P. ostreatus

Protein concentration in the fruit body was highest when mushrooms were cultivated on *C. sophera* and *P. argentatum* (Table 3). However, supplementation of those two

Table 3
Protein values of *Pleurotus ostreatus* fruit bodies in two flushes grown on non-heat treated rice straw and weed species and harvested after three days of primordia formation

| Substrate | Protein mg/g of fresh fruit body | | | |
|---|---|---|---|---|
| | Mean | Std. Deviation | *p*-Value | Statistical rank |
| *Cassia sophera* | 10.85 | 0.96 | | A |
| *Parthenium argentatum* | 10.13 | 1.05 | >0.05 | A |
| *Leonotis* sp. | 8.44 | 0.82 | <0.05 | B |
| Rice straw + *Leonotis* sp. | 7.52 | 0.12 | <0.05 | C |
| *Ageratum conyzoides* | 7.23 | 0.84 | <0.05 | D |
| *Lantana camara* | 6.94 | 0.73 | >0.05 | D |
| Rice straw | 6.7 | 0.82 | >0.05 | D |
| Rice straw + *Ageratum conyzoides* | 6.52 | 0.59 | >0.05 | D |
| Rice straw + *Sida acuta* | 6.34 | 0.23 | <0.05 | E |
| Rice straw + *Cassia sophera* | 6.09 | 0.68 | <0.05 | F |
| Rice straw + *Tephrosia purpurea* | 6.08 | 0.58 | >0.05 | F |
| *Sida acuta* | 5.8 | 0.68 | >0.05 | F |
| Rice straw + *Lantana camara* | 5.79 | 0.42 | >0.05 | F |
| Rice straw + *Parthenium argentatum* | 5.75 | 0.6 | >0.05 | F |
| *Tephrosia purpurea* | 5.2 | 0.72 | <0.05 | G |

weeds with rice straw resulted in decrease in protein level of the fruit bodies. The protein concentration of the fruit bodies derived from *Leonotis* sp. singly showed better results than the rice supplemented weeds. Except *T. purpurea*, the protein content of the fruit bodies obtained from different weeds were better than that obtained from rice straw supplemented weeds (Table 3).

## 4. Conclusion

Selective weeds can be used successfully as substrates for oyster mushroom cultivation. Weeds are not only proved as the alternative substrate for oyster mushroom cultivation, they also can significantly increase the protein content and reduce the production time. Supplementation of weed substrate with rice straw increases the accumulated biological efficiency of mushroom, stimulating mainly the production in the second flush. In the present investigation *Leonotis* sp. has been identified as the best substrate for oyster mushroom cultivation with respect to biological efficiency and fruiting time. Therefore, oyster mushroom cultivation proves to be a highly efficient method for disposing of weed plants as well as producing protein-rich food.

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