

Cultivation and Proximate Analysis of Pink Oyster Mushroom (*Pleurotus eous*) in Doimukh, Arunachal Pradesh

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Abstract

Pleurotus eous also known as Pink oyster mushroom is an edible mushroom species and regarded as food because of their high nutritional values with a rich source of dietary fibers, protein and minerals and exclusively low fat contents. The cultivation of *Pleurotus eous* was carried out during the month of May - June, 2022 at the mushroom hut built in the premises of Botany department, Rajiv Gandhi University, Arunachal Pradesh and the nutrient composition of harvested fruitbodies of *Pleurotus eous* grown on various substrates such as Paddy straw (PS), Rice Bran (RB), *Saccharum spontaneum* (SS), PS+SS, PS+RB and SS+RB were determined according to the standard protocol. The nutritional component of all the selected substrates except Rice bran (contaminated) supported the growth of *Pleurotus eous*. The mushrooms harvested from SS+RB contained the highest moisture (93.65%), ash (10.11%), and crude fibre (28.01%), while the highest protein contents (29.0%) was observed in PS+SS. It has been observed that cultivated mushrooms, *Pleurotus eous* are abundantly endowed with nutritive constituents, which could be used as source of food and medicine as they are rich in protein and low in fats.

Keywords: Food, Mushrooms, Nutritional, *Pleurotus eous*

INTRODUCTION

Oyster mushrooms (*Pleurotus* spp.) are group of edible mushrooms that naturally grow on decayed wooden logs or sometimes on tree trunks in found in tropical and temperate forests^[1]. They are commercially cultivated worldwide especially in Asia, Europe, Africa and

in India, because of their fast growth, high yield, less preparation time, minimum space and low economic investment in their cultivation and presently ranks as second highest cultivated edible mushrooms after button mushrooms^[2]. Oyster mushrooms (*Pleurotus* spp.) have been reported to have high nutritional and health values with rich source of proteins, dietary fibres, high K:Na ratio, vitamin content and micro and macro-elements, carbohydrates and low fats and cholesterol value which makes them good for the patients with hypertension, diabetes and heart diseases^[3,4]. The comparatively low contents of starch and fats in oyster mushrooms are supported by findings of Kalogeropoulos *et al.*^[5] who reported low levels of fat vegetables and carbohydrate in mushrooms. Large amount of macromolecules compounds like polysaccharide-peptides and polysaccharide- protein complex have been reported in these mushrooms and found to have culinary values such as antimicrobial, antihypercholesterolemic, antioxidant, antidiabetic, anticancer, and immunomodulatory properties^[6,7].

Pink oyster mushroom (*Pleurotus eous*) are popularly cultivated for their good flavor, aesthetic appeal, texture and in comparison to other commercially cultivated oyster mushrooms, *Pleurotus eous* gave higher biological efficiency and has shorter life cycle. The cultivation of mushrooms needed a suitable substrates that consist of organic and inorganic materials to complete their life cycle, they are highly specific and not only serve as a physical support but also as good source of nutrients for their growth and development^[8].

Pleurotus eous are reported to widely cultivated on various agricultural waste materials such as banana pseudo stem, dry fruit of ridge gourd, mustard stem, groundnut shell, maize cob, rice husk, pea straw, tobacco stem and paddy straw^[9], coir pith, bamboo leaves, cotton stalks, pearl millet stalks, palm leaves, groundnut haulms, soybean stalks, sorghum stalks, sugarcane bagasse and trash, weeds and tapioca leaves^[10]. Like many other *Pleurotus* spp., *Pleurotus eous* also reported to have nutritional and medicinal properties. Nutritional analysis of *Pleurotus eous* reported that it contained crude fiber (12%), protein content (46%), and relatively low fat content (1.2%)^[11], while other reported protein with value of 30.50%, crude fiber (9.00%) and fat (2.62%)^[12]. The current study was conducted to examine the nutrient composition of initial substrates and to investigate the effects of selected lignocellulosic substrates on the nutrition constituents of produced mushrooms and the spent mushroom substrates on which they were grown.

MATERIALS AND METHODS

Cultivation of *Pleurotus eous* was conducted in the mushroom hut built in the premises of Department of Botany, Rajiv Gandhi University, Arunachal Pradesh and the nutrition analysis was done in the laboratory of G. B. Pant National Institute of Himalayan Environment and Development North-East Unit, Itanagar, Arunachal Pradesh.

Mushroom species and spawn preparation

The selected mushroom strain, *Pleurotus eous* (Berk.) Sacc. was procured from the culture bank of Directorate of Mushroom Research (DMR), Solan, Himachal Pradesh and the mother

culture was maintained in Potato Dextrose Agar (PDA) at $25\pm 2^{\circ}\text{C}$ in B.O.D incubator. Whole wheat grain was used for spawn preparation and the grains were filled in autoclavable polypropylene bags and autoclaved at 121°C for 2 hours. The sterilized bags were inoculated with selected mushroom strain and kept at $25 \pm 1^{\circ}\text{C}$ for 15 days^[13].

Substrate preparation and mushroom cultivation

Three different types of lignocellulosic materials and their combination were used as substrates for growing *Pleurotus eous* such as Paddy straw (PS), Rice Bran (RB), *Saccharum spontaneum* (SS), PS+SS, PS+RB, and SS+RB in the combination ratio of 100%, 100%, 100%, 1:1, 1:1 and 1:1 on dry weight basis respectively. The substrates were dried and chopped into small pieces (2-4 cm) and mixed at different ratio (on dry weight basis). The substrates were sterilized by autoclaved at 121°C for 15 mins^[14] and then inoculated with 3% (w/w) grain spawn and incubated at $20-27^{\circ}\text{C}$ under dark conditions in mushroom hut for mycelial colonization. After complete mycelial colonization, these bags were opened for primordial formation and environmental parameters such as relative humidity (73% to 96%), light and aeration were maintained. Fresh and matured mushrooms were harvested manually by hand without disturbing the mushroom beds at one time for uniform harvesting to the next flush.

Proximate composition of the harvested mushrooms, *Pleurotus eous*

Before proximate analysis harvested mushrooms were dried at 60°C in oven until constant weight was obtained and grounded into powder for further nutrient analysis^[15].

Moisture content determination^[16]

A clean and dry crucible was weighed (W1) and two grams of the samples were transferred into the crucible and weighed (W2) and then kept into hot air oven at 150°C for 2 h until constant weight (W3) was obtained. The moisture content (%) was calculated by using the formula:

$$\% \text{ Moisture} = \frac{W2 - W3}{W2 - W1} \times 100$$

Where;

W1 = Initial weight of clean & dry crucible; W2 = Initial weight of crucible + fresh sample

W3 = Final weight of crucible + sample after drying

Fats content determination^[16]

A clean and dry boiling flask of 300 ml was dried at 100°C in an oven and weighed (W2). 1 g of the sample was weighed (W1) and put into the thimble. The thimble with samples were transferred into the soxhlet apparatus containing the solvent, petroleum ether and the extraction was done for atleast 6 to 8 h. After extraction was completed, the flask containing the oil content was dried at 80°C for 30 mins in an oven and cooled in a desiccator and final weighed (W3) was taken. The fat content was calculated as follows:

$$\% \text{ Crude fat} = \frac{W3 - W2}{W1} \times 100$$

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Where;

W1 = Weight of sample; W2= Weight of empty flask; W3 = Weight of flask + fat

Ash content determination ^[17]

Crucibles were dried properly in an oven and cooled in a desiccators and weighed (W1). 2 g (W2) of the samples in crucible were transferred into the muffle furnace and kept for atleast 550°C for 4 h. The crucibles were removed from muffle furnace and again cooled in a desiccators and the final weight was taken as W3. The ash contents were obtained as follows:

$$\% \text{ ash} = \frac{W3 - W1}{W2} \times 100$$

Where;

W1= Weight of crucible before ashing; W2= Weight of sample; W3= weight of crucible after ashing

Crude protein determination ^[16]

0.2 g of the sample was transferred into Kjeldahl flask containing mixture of 3g of hydrated cupric sulphate + 20 ml sodium sulphate and 1 g of Conc. H₂SO₄ (N x 6.25) to digest the sample. After digestion was completed the solution became clear and was cooled properly and then filled the solution with distilled water up to level of 100ml. 5 ml of digest was collected for distillation process and then 5 ml of NaOH was transferred into the distillation flask and then distilled for sometimes. The indicator, Boric Acid absorbed the liberated ammonia from distillation unit and this was finally titrated with standard acid, HCL until the solution colour changes to pink from green colour. The crude protein content was determined by using the formula:

$$\% \text{ crude protein} = \frac{T \times S \times 0.01}{A \times W} \times 6.25$$

Where:

T = Titre value; S = Final digest solution; A = Aliquot volume; W = Sample weight

Crude fibre determination ^[16]

1g of the sample (W1) was boiled with 200 ml of a solution having 2.50 g of H₂SO₄ and then using a cotton cloth the solution was filtered and the residue obtained was again boiled with 200 ml of solution filled with 2.50 g of NAOH for another 30 mins. The final residue obtained was then again filtered through cotton cloth and transferred into oven for drying and weighed (W2). The final residue formed was then converted into ashes in a muffle furnace and cooled in desiccator and weighed (W3);

$$\% \text{ Crude fibre} = \frac{W2 - W3}{W1} \times 100$$

Where:

W1 = Weight of the sample

W2= Weight of the sample after drying in an oven

W3 = Weight of the sample after ashing in a muffle furnace

Total Carbohydrate determination^[17]

The total carbohydrate was determined by using the formula

Total carbohydrates (%) (dry weight basis)

= 100 – [moisture (%) + crude fibre (%) + protein (%) + ash (%) + fat (%).

Data analysis

All the experiments were carried out in three replicates (n=3) and the results were given as the mean ± standard deviation. All the data of the experiment were subjected to one way ANOVA and means were compared with least significant difference (LSD) at 5% probability level using SPSS, Version 18.

RESULTS AND DISCUSSION

Proximate composition of the harvested mushrooms, *Pleurotus eous*

Proximate composition of the harvested mushrooms from different substrates has been shown in Table 2. The moisture content of the fresh mushrooms were found in the ranged of 88.34 – 93.65%. The values obtained in this study are similar to the reported values of 90.97-91.98% moisture content from *Pleurotus eous* cultivated on various agricultural waste materials studied by Sathyaprabha and Panneerselvam^[18]. In the current study the harvested mushroom from SS contained the highest fats content with value of 1.42% and the lowest was found in PS with a value of 1.04%. Ingale and Ramteke¹⁹ also reported low fat content of 1.2 - 1.9% in various *Pleurotus* spp. Low fats contents ranged from 1.0-4.3% were obtained from some edible mushrooms by Valverde *et al.*^[19]. In this study, the ash content of the harvested mushrooms were ranged from 7.94 -10.11%, which is in consistent with the findings of Jin *et al.*^[20] who reported ash contents of 7.82-9.65% in oyster mushroom, *Pleurotus ostreatus* grown on various agro-wastes. The highest protein content (29.0%) was found in *Pleurotus eous* harvested from PS+SS whereas lowest in SS (11.50%). The obtained protein contents (11.50 to 29.0%) are in agreement with the studies performed by FAMILONI *et al.*^[21]. According to Gupta *et al.*^[22] the protein contents in *Pleurotus sajor-caju* were found in the ranged of 27.4–34%, the researchers suggested that the variation in the protein contents were due to the influence of types of substrates. *Pleurotus eous* harvested from SS+RB had the highest crude fibre contents (28.01%) whereas lowest crude fibre content of 15.32% was observed in PS+SS. The obtained value is in consistent with the reported value of 21.8- 27.4% crude fibre contents from varieties of oyster mushroom species cultivated in Bangladesh by Ahmed *et al.*^[23] but greater than 11.4 to 13.3% crude fibre contents reported from

Pleurotus sajor-caju studied by Chang *et al.*^[24]. The highest carbohydrate contents were found in the *Pleurotus eous* harvested from SS (63.0%) while the lowest value of 44.02% was observed in fruitbodies harvested from SS+RB. Telang *et al.*^[25] reported total carbohydrate contents of value ranged from 49.0 to 52.0% while evaluating nutrient composition of *Pleurotus eous*. The study showed that the type of substrates had a variable influence on the nutrient composition of harvested *Pleurotus eous* which is in agreement with the findings of Maftoun *et al.*^[25]

Table 2. Proximate composition (% dry weight basis) of the fruitbodies of *Pleurotus eous* harvested from various substrates (PS: paddy straw; RB: rice bran; SS: *Saccharum spontaneum*)

Substrate	Moisture	Fats	Ash	Protein	Crude fibre	Total carbohydrate
PS	88.34 ^b ± 0.52	1.04 ^a ±0.01	7.94 ^{bc} ±2.33	14.81 ^{ac} ±3.3	15.50 ^b ±0.51	60.02 ^{ab} ±5.45
RB	Contaminated	-	-	-	-	-
SS	91.11 ^c ± 0.16	1.42 ^a ±0.50	8.05 ^c ±0.07	11.50 ^a ±2.45	16.22 ^b ±1.01	63.0 ^b ± 2.00
PS+RB	89.32 ^a ± 0.52	1.11 ^a ±0.07	8.21 ^c ±0.21	16.40 ^{ac} ±4.71	21.33 ^b ±0.59	52.94 ^c ±4.14
PS+SS	90.0 ^d ± 0.42	1.30 ^a ±0.20	8.72 ^b ±0.46	29.0 ^b ±1.89	15.32 ^c ±0.65	45.66 ^c ± 1.94
SS+RB	93.65 ^e ±0.54	1.12 ^a ±0.08	10.1 ^b ±0.16	16.72 ^c ±0.55	28.01 ^d ±0.96	44.02 ^d ±1.53

Mean are expressed as % of dry weight basis except moisture, which is expressed as % of fresh weight. The results are the means ± SD of 3 replicates (n=3), Means with same letters within same column are not significantly different at (P>0.05)

CONCLUSION

The study showed that the nutritional constituents of various lignocellulosic substrates supported the growth of pink oyster mushroom, *Pleurotus eous*. The proximate analysis of harvested mushrooms showed variable ranged in nutritional composition such as moisture, fats, ash, protein and total carbohydrates. Generally mushrooms are rich in protein and low in fats, in this study the mushrooms grown on the substrate, PS+SS showed the highest protein contents. The fats contents in the mushrooms harvested from various substrates was found in a very low amount which ranged from 1.04 – 1.42 %. Thus *Pleurotus eous* mushrooms could be considered as food supplements in diet with high nutrients value and hence helps to eliminate malnutrition.

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