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Cultivation of oyster mushroom [*Pleurotus ostreatus* (Jacq.) P. Kumm] through substrate enriched with tea waste

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ABSTRACT

A study was conducted to assess yield, biological efficiency and nutritional contents of *Pleurotus ostreatus* on different formulations of wheat straw, sugarcane bagasse, coconut green husk, banana pseudostem and waste paper mixed with tea waste, lime, gypsum and sucrose at various proportion. The data showed the maximum yield of *Pleurotus ostreatus* on wheat straw + tea waste + lime + gypsum + sucrose (711.6 g/kg substrates) and sugarcane bagasse + tea waste + lime + gypsum + sucrose (702.3 g/kg substrates). The nutritional contents of the mushroom grown on mixed substrates were comparatively higher as compared to control. Mix substrates gave the maximum protein content (27.316 mg/g), total sugar (27.670 mg/g), phenol (1.952 mg/g), Moisture (91.6 %).

Keywords: Oyster mushroom, *Pleurotus ostreatus*, mix substrate, yield, nutrition

The cultivated *Pleurotus* mushroom includes a number of different species: *P. ostreatus, P. florida, P. eryngii, P. cystidiosis, P. flabellatus, P. cornucopie,* and *P. sajor-caju. Pleurotus* mushroom are wide spread in the temperate zone representing the third most popularly grown mushroom in the world and rank second in India. The word mushroom is derived from the Gallo Roman *mussiro*, which is defined as a "macro-fungus" that has a fleshy and used throughout the world to describe the fruiting bodies of saprophytic, mycorrhizal and parasitic fungi. It is either epigenous or hypogenous and large enough to be seen with the naked eye and to be picked by hand (Chang and Miles,1992). The cultivated Oyster mushroom (*Pleurotus ostreatus*) mostly belongs to the family Agaricaceae of class Basidiomycetes (Alexopoulos *et al.*, 2012). It is popularly known as 'Dhingri' in India and grow naturally in the temperate and tropical forest on dead and decaying wooden logs or sometimes on dying trunks of deciduous or coniferous woods. It may also grow on decaying organic matter.

Oyster mushroom is cultivated using various agricultural wastes. The appropriate preparation of the substrate is crucial for enhanced yield of mushrooms. Mushroom's substrate may be simply defined as a kind of lignocellulosic materials, which supports the growth, development and fruiting of mushroom mycelium. Lignocellulosic materials such as cereal straws, dried leaves, banana leaves, etc. act as suitable base substrates for mushroom production (Chadha and Sharma,1995). Indian diet is primarily based on cereals (wheat, rice and maize), which is deficient in protein. Supplementation of mushroom recipe in Indian diet will bridge protein gap and improve the general health of socio-economically backward communities (Manikandan, 2011). Popularity of Oyster mushroom

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has been increasing due to its simple and short substrate preparation, cultivation process, high yield potential and high nutritional value.

Oyster mushroom can grow at moderate temperature that ranges from 20 – 30°C and relative humidity 55 – 70 per cent for a period of 6-8 months in a year. It can also be cultivated in summer season by providing the extra humidity needed for its growth. Thus, this mushroom can be a productive vocation for landless farmers and women to augment their income using locally available substrate. This study is aimed to formulate a locally available productive substrate combination to grow oyster mushroom with a good biological efficiency and to see the effect of substrate composition on their nutritional value.

MATERIALS AND METHODS

Study area and experimental materials

The experiment was conducted at the Department of Plant Pathology, College of Agriculture, Junagadh Agricultural University, Junagadh during 2020-21. Wheat straw, sugarcane bagasse, banana pseudostem, coconut green husk and waste paper were gathered from farmer's field of Saurashtra region. The tea waste collected from residue left after the water-soluble contents of tea had been extracted in hot water. Gypsum, lime, sucrose and pure culture of *Pleurotus* ostreatus. was collected from mushroom production unit of Junagadh Agricultural University. Experiment was conducted using Completely Randomized Design for checking the morphological and biochemical parameters of difference in different substrates.

Growth and pure culture of mushroom

Potato dextrose agar (PDA) medium was poured in sterilized Petri plate and inoculated with 10 mm disc of *P. ostreatus* under aseptic condition. Then, the Petri plate were incubated at 25 ± 2°C. The diameter of colony was measured after 6 days of growth. Sub culturing was done on PDA slants as and when needed.

Preparation of spawn

Spawn was prepared by following the standard method on sorghum grain. The sorghum was washed to remove dirt, dust and also sorted to remove unwanted materials like stone, metals and broken grains. The sorted sorghum was soaked in clean water and steamed for 30-45 minutes. Antibiotic chloramphenicol was added (1 capsule 250 mg was added per 1 kg of grain). Calcium carbonate was mixed with grains to maintain the pH, filled in 300g in each bag, and sterilized in autoclave for 30 minutes at 121°C with 15 lbs pressure. The bags were inoculated with master culture and were incubated until the mycelium completely colonized the sorghum seeds.

Substrate preparation and spawning

The basic substrate used in the study were wheat straw, sugarcane bagasse, banana pseudostem, coconut green husk and waste paper while the mix substrates have a combination of tea waste, gypsum, sucrose and lime. Each substrate was taken separately (around 12 kg) in a plastic tub. The substrates were sterilized chemically by soaking with carbendazim 50% WP @ 10 g/10 liters of water and 40% of formaldehyde solution (13.50 ml/10 liters of water) for a period of 24 hours. Tea waste was sterilized in autoclave. In case of office waste, small pieces of paper and carton were wetted to make 65% moisture level. The bags were sterilized in autoclave. After the sterilization, papers and carton pieces used as a substrate.

The substrate combination used were T-1: Coconut green husk + tea waste + lime + gypsum +

sucrose (70:27:1:1:1), T-2: Sugarcane bagasse + tea waste + lime + gypsum + sucrose (70:27:1:1:1), T-3: Banana pseudostem + tea waste + lime + gypsum + sucrose (70:27:1:1:1), T-4: Waste paper + tea waste + lime + gypsum + sucrose (70:27:1:1:1), and T-5: Wheat straw + tea waste + lime + gypsum + sucrose (70:27:1:1:1). Wheat straw (T-6) alone was used as control.

Data collection and analysis

The experiment was carried out to find the effective substrate for agronomic traits. Data were recorded on five various substrates supplemented with tea waste, gypsum, lime and sucrose for total number of fruiting bodies, height of fruiting bodies (cm), diameter of stipe (cm), stipe length (cm), pileus diameter (cm), yield (gm/kg) and biological efficiency (%) using the procedure suggested by Gopinath (2015).

Biological efficiency of mushroom was calculated by using formula as recommended by Chang and Miles (1989). Biological efficiency was calculated as the ratio between the fresh weight of mushrooms and the dry weight of substrate per bag expressed in percentage.

$$\text{Biological efficiency (\%)} = \frac{\text{Fresh weight of mushroom (g)}}{\text{Dry weight of substrate (g)}} \times 100$$

Biochemical Analysis

The various biochemical analysis of fruiting body of *Pleurotus ostreatus* harvested from different substrates was done for different biochemical parameter (protein, total sugar, phenol, carbohydrate and moisture) by standard biochemical procedures. Protein content was measured by Folin-Lowry's method (Lowry *et al.,* 1951), Total sugar was analysed by phenol sulphuric acid method (Dubois *et al*., 1956). Total phenol by phenol-folin method (Snell and Snell, 1953), Moisture content by method of Gaur *et al*. (2016) and Carbohydrate content by Dinitrosalicyclic acid (DNS) Method (Miller, 1959).

RESULTS

Yield and yield related parameters

Rate of growth of *P. ostreatus* was studied on the four various supplemented substrates as well as wheat straw substrates as a control. Data (Table 1) showed good yields in the substrates added with supplements like tea waste, gypsum, sucrose and lime as compared to the wheat straw (control). The maximum number of fruiting body (36 no.) was observed in wheat straw T_5 and sugarcane bagasse T_2 with combination of tea waste, lime, gypsum and sucrose.

With respect to height of the fruit body, supplemented wheat straw T_5 and sugarcane bagasse T_2 recorded maximum height. Also, the maximum stipe length was achieved in wheat straw combination T_5 (7.20 cm), stipe diameter (2.20 cm) and pileus diameter (11.20 cm) which was statistically at par with sugarcane bagasse combination T_2.

The fresh weight of mushroom varied significantly with the substrates. In the present study, treatment T_5 and T_2 gave the maximum yield (711.60 g/kg and 702.30 g/kg dry substrate, respectively). Highest and lowest dry weight of mushroom was found in supplemented sugarcane bagasse T2 (11.10 g/100g) and banana pseudostem T3 (8.50 g/100 g), respectively.

Maximum per cent biological efficiency was observed in supplemented wheat straw T_5 (71.16 %) followed by sugarcane bagasse T_2 (70.23 %). The study concluded that among different combinations, wheat straw + tea waste + lime + gypsum + sucrose (70:27:1:1:1) was found superior among all the different supplemented treatments.

Table 1. Effect of different substrates on growth of mushroom

Sr. No.	Substrate mixture	Total no. of fruiting	Height of fruiting body body	Stipe length (cm) (cm)	Stipe diameter (cm)	Pileus diameter (cm)	Yield (g/kg)	Total dry weight of mushroom	Biological efficiency (%) (g/100 g)
T_1	Coconut green husk + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	34	14.0	6.8	2.0	10.9	689.0	10.6	68.90
T_2	Sugarcane bagasse + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	36	14.5	7.0	2.1	11.0	702.3	11.1	70.23
T_3	Banana pseudostem + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	33	13.0	6.7	1.8	10.8	645.2	8.5	64.52
T_4	Waste paper + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	26	8.9	5.5	1.8	7.6	453.5	9.7	45.35
T_5	Wheat straw + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	36	15.0	7.2	2.2	11.2	711.6	9.4	71.16
T_6	Control (Wheat straw)	32	12.3	6.4	1.8	10.7	611.9	8.9	61.19
	S.Em.±	0.91	0.26	0.08	0.04	0.10	11.48	0.16	1.14
	CD at 5 %	2.81	0.81	0.26	0.12	0.33	35.40	0.50	3.54
	CV %	4.82	3.52	2.23	3.63	1.80	3.13	2.95	3.13

Nutritional quality of *Pleurotus ostreatus* grown on mix substrates

The biochemical content of the mushroom (*Pleurotus ostreatus*) harvested from the various mix substrates are shown in Table 2. Wheat straw, sugarcane bagasse, coconut green husk, banana pseudostem and waste paper were used with combination of tea waste, lime, gypsum and sucrose in different proportion to analyse the biochemical properties of fruit bodies i.e. total protein, phenol, total sugar, moisture and carbohydrate.

The nutritional contents of the mushroom grown on mixed substrate were comparatively higher as compared to that of single substrates (control). Nutritional analysis of oyster mushroom showed total protein ranged from 19.80 to 27.31 mg/g, total sugar from 4.71 to 27.67 mg/g, moisture content from 88.90 to 91.60 % and carbohydrate from 2.64 to 8.91 mg/g.

The maximum total protein content (27.31 mg/g) was obtained from wheat straw combination T_5 followed by supplemented coconut green husk T_1 (25.57 mg/g). The supplemented waste paper T_4 was found least effective having protein content (19.80 mg/g) among other treatment. The supplemented wheat straw T_5 was found being superior among the different substrates to indicate maximum content of total sugar (27.67 mg/g). Lowest content of sugar (4.71 mg/g) found in sugarcane bagasse combination T_2. Among various mix substrates wheat straw T_5 gave maximum phenol content (1.95 mg/g) as compared to the other substrate. Lowest phenol content (1.53 mg/g) obtained from waste paper combine with supplements T_4.

Table 2. Effect of different substrates on nutritional composition of mushroom

Sr. No.	Substrates mixture	Protein (mg/g)	Total Sugar (mg/g)	Phenol (mg/g)	Moisture (%)	Carbohydrate (mg/g)
T_1	Coconut green husk + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	25.575	17.703	1.624	89.4	2.642
T_2	Sugarcane bagasse + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	23.150	4.715	1.866	88.9	8.911
T_3	Banana pseudostem + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	24.072	14.254	1.741	91.5	4.766
T_4	Waste paper + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	19.804	19.816	1.535	90.3	4.663
T_5	Wheat straw + tea waste + lime + gypsum + sucrose (70:27:1:1:1)	27.316	27.670	1.952	91.6	5.958
T_6	Control (Wheat straw)	25.600	22.545	1.910	91.0	5.022
	S.Em.±	0.366	0.457	0.035	0.54	0.106
	CD at 5 %	1.128	1.410	0.109	1.68	0.327
	CV %	2.580	4.070	3.470	1.05	3.450

Highest moisture percentage was observed in mushroom harvested from mix substrate wheat straw T_5 (91.60 %) and banana pseudostem T_3 (91.50 %). Whereas, lowest moisture percentage was found in mushroom obtained from sugarcane bagasse used as a mix substrate (88.90 %). The maximum amount of carbohydrate (8.91 mg/g) was obtained from sugarcane bagasse supplemented T_2 followed by supplemented wheat straw T_5 (5.95 mg/g) substrate. In case of the supplemented banana pseudostem T_3 and waste paper T_4 (4.76 and 4.66 mg/g) both were statistically at par with each other. Whereas, lowest amount of carbohydrate obtained from supplemented coconut green husk T_1 (2.64 mg/g).

DISCUSSION

Wheat straw is one of the easily available agriculture wastes and hence it is taken as control to check the effect of different supplements added to substrate. Five substrates *viz*., wheat straw, sugarcane bagasse, coconut green husk, banana pseudostem and waste paper were evaluated in combination with tea waste, lime, gypsum and sucrose (70:27:1:1:1) and wheat straw alone served as control for yield and yield related parameters.

Results of the present study are similar with findings of Pathania *et al.* (2017), Garg (2013) and Mohamed *et al*. (2016). Tirkey *et al*. (2017) determined the best effective substrates that support the maximum yield with the highest biological efficiency and nutritional contents. They concluded that wheat straw and banana leaves substrates individual as well as in combination proved to be the best for cultivation of oyster mushroom. The results were also found lined in with Dunder *et al.* (2008), Fanadzo *et al*. (2009) and Khan *et al.* (2012). Stanley *et al*. (2011) reported the supplemented (mix) substrates gave better result in morphological characteristics than un-supplemented (single) substrates.

Chukwurah *et al*. (2013) studied the correlation of stipe length, pileus width and stipe girth of oyster mushroom. They reported the highest biological efficiency (97.9%) in the substrate composed of two agricultural wastes. Present results were also

supported by finding of Rukhsana *et al.* (2019). They reported that lime and gypsum acts as a buffer for the mycelial expansion of oyster mushroom. So, based on the results obtained in this experiment. The results are in consonant with the report of Ogundele *et al*. (2014) who also reported that the total protein content, ash, crude fiber and crude fat were higher in the mushroom harvested from mixed substrate than that of the pure substrates.

CONCLUSION

Among different treatment maximum yield was obtained from wheat straw + tea waste + lime + gypsum + sucrose (70:27:1:1:1) (711.60 g/kg substrates) which was found superior among all substrates. While, waste paper was least productive in terms of yield of mushroom when used as substrate either singly or as mixed substrate. Mix substrates gave the maximum protein content (27.31 mg/g), total sugar (27.67 mg/g), phenol (1.95 mg/g) in wheat straw + tea waste + lime + gypsum + sucrose and carbohydrate content (8.91 mg/g) in sugarcane bagasse + tea waste + lime + gypsum + sucrose. However, the moisture content was reported higher in pure substrate (92.10 %) as compared to mixed substrates (91.60 %).

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