

EFFECT OF DIFFERENT FLOURS WITH BAGGASE ON YIELD OF *PLEUROTUS EOUS*

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ABSTRACT

Mushroom supplementation consists of the application of nutritional amendments to the substrates employed for mushroom cultivation. Sugarcane bagasse was supplemented with different flours viz., bajra, chick pea, wheat, green gram, jowar as a substantial impact on mushroom yield and quality. All substrates gave maximum mycelium growth of *Pleurotus eous* at 15 days of incubation period as compared to 5 and 10 days. On the other hand, sugarcane bagasse along with all mixed flours gave maximum yield of *Pleurotus eous* followed by wheat, green gram, chick pea, bajra and jowar.

Key words: Mushroom, sugarcane bagasse, nutritional substrates, *Pleurotus eous*.

INTRUDUCTION

Sugar industry is the second largest industry in India, which discharges large quantities of liquid and solid wastes. The sugarcane residue, the bagasse, is one of the most abundant agro-industry by-products in the world, producing about 540 million tons of residues per year (Satyanarayana et al., 2009). It is generally composed of approximately 40% cellulose, 24% hemicelluloses, and 25% lignin (Shah, 2003) and small amounts of ash and waxes. Bagasse is the fibrous residue that remains as a waste product from the sugar milling process, and is often used to fuel boilers at sugar mills (Visvanathan et al., 1998). Sugarcane bagasse is a residue produced in large quantities by sugar industries. In general, one metric ton of sugarcane bagasse generated 280kg of bagasse, the fiberous byproduct remaining after sugar extraction from sugar cane (Sun et al., 2004).

Mushroom cultivation on sugarcane bagasse is also practiced in some countries. Agro-waste can be managed through mushroom cultivation, since mushrooms have the ability to grow on a variety of raw lignocellulosic substrates and under a wide range of temperatures (Sanchez, 2010). White-rot fungi could be considered a very efficient tool for the biological degradation of lignin through the microorganisms' action. For this purpose, Dong, et al. (2013) studied the lignocellulosic degradation process of sugarcane bagasse by means of different white-rot fungi (*P. chrysosporium*, *Lentinula edodes*, and *P. ostreatus*). Ritota and Manzi (2019) reported that, oyster mushroom has biodegradation capability and nutritional value. *Pleurotus* spp. could be a useful tool to reduce the environmental impact of agri-food wastes and to transform them into new resources for the simultaneous production of edible food items with high added value.

Cultivation of mushrooms using agricultural and Industrial residues provides a very cheap and ecofriendly alternative for producing foods with high nutritional value. Currently, about 40% of sugarcane bagasse from sugar factories and a large quantity of wastepaper as well as biomass from lignocellulosic plants, which could have been used as cheap source for mushrooms growth.

The cultivation of oyster mushroom requires the use of cellulosic materials or residues. These residues and byproducts can be recovered and upgraded to higher value and useful products by using them as growth substrates (Dawit, 1998). It might be the only current process that combines the production of protein-rich food with the reduction of environmental pollution (Beetz and Kustudia 2004). The production of mushrooms is regarded as the second most important commercial microbial technology next to yeast (Pathak et al. 2009). Mushrooms have been eaten and appreciated for their flavor, economic and ecological values, and medicinal properties for many years. In general, mushrooms contain 90% water and 10% dry matter (Morais et al. 2000; Sánchez 2004). Mushrooms are being used as food and medicine since time immemorial. According to Chang (1992) the protein value of dried mushroom has been found to be 30-40% containing all the essential amino acids. Mushrooms constitute an ideal source to reduce body weight. Mushroom helps to reduce serum cholesterol and high blood pressure (Mori et al., 1986). Mushrooms are being used as food and medicine since time immemorial. According to Chang (1992) the protein value of dried mushroom has been found to be 30-40% containing all the essential amino acids. Mushrooms constitute an ideal source to reduce body weight. Mushrooms supply more protein per unit area than other crops (Gupta, 1986). *Pleurotus* sp. is popular and widely cultivated throughout the world mostly in Asia and Europe owing to their simple and low cost production technology and higher biological efficiency (Mane et al., 2007). *Pleurotus* species, commonly known as oyster mushrooms are a group of higher fleshy fungi belonging to the Basidiomycetes. They are considered as one of the four major edible mushrooms cultivated in different countries for human consumption. *Pleurotus* spp. are selective degraders, degrading lignin and hemicellulose rather than cellulose. Furthermore, cultivation of *Pleurotus* spp. is very simple compared to that of the most commonly cultivated mushroom, *Agaricus bisporus* (Cohen, et. al., 2002). For *Pleurotus* cultivation, non-composted, chopped and water-soaked straw is sufficient (Philippoussis, 2009). The present study was initiated to investigate the suitability of various organic flours such as bajra, chick pea, wheat, green gram, jowar with bagasse on growth performance and yield of oyster mushroom. The implication of this study is to facilitate technology adoption of oyster mushroom cultivation using sugarcane bagasse wastes along with different flours and thereby identify the feasibility of mushroom cultivation in the study area for the betterment of the life of the local community. The results of the study will be crucial to poor rural communities which utilizes bagasse for *Pleurotus* eous mushroom production for both food and income generation.

MATERIAL AND METHODS

Spawn Preparation

The term 'spawn' refers to cereal kernels overgrown by mushroom mycelium. Spawn is used as "seed" for inoculating substrates with the mushroom mycelium. Spawning was carried out aseptically, preferably using the same transfer chamber or the same inoculation room as was used in spawn preparation. Grain or sawdust spawn was used to inoculate the substrate in bags. With grain spawn, the bottle is shaken to separate the seeds colonized with the white mycelium. After lifting the plug and flaming the mouth of the bottle, a few spawn grains (about 1 to 2 tsp.) were poured into the substrate bag. Both the plug of the spawn and the plug of the compost bag were replaced and the next bags were then inoculated. The newly inoculated bags were slightly tilted to distribute the grains evenly in the shoulder area of the bag around the neck. For sawdust spawn, the spawn was broken up with an aseptic needle. A piece of the spawn then transferred by using a long flat-spooned needle especially designed to scoop the spawn. One bottle of grain or sawdust spawn in a 500 ml dextrose bottle was sufficient to inoculate 40 to 50 bags.

Sterilization and preparation of substrates

For cultivation of mushroom substrates, viz. sugarcane bagasse they were sterilized as follows: These sugar industrial waste were soaked in water for 12 hours before use to soften the tissues. Wastes were chemically sterilized in plastic pots as shown in plate No. 1. 25 liters of water was taken in a plastic pots. 5kg waste slowly steeped in water. In another plastic bucket, carbendazim 50% WP (75.ppm), Bavistin 7.5.g and 25 ml formaldehyde (37- 40%) were dissolved and slowly poured on the already soaked bagasse straw. Straw was pressed and covered with a polythene sheet. After 24 h straw was taken out and excess water drained. The lab was fumigated with the chemical formaline and KMnO₄. The preferable room temperature 26 ± 2°C was kept with relative humidity of 70 – 80%.

Bag filling Method: flours add + baggase

Bag filling method was used throughout the studies. The polythene bags of 14 x 22 cm and the bottom of the bags were tied with a rubber to provide a flat circular bottom to the mushroom beds. Dry weight of the substrates was recorded and the bags full of different substrates were weighed and were maintained at 2.5 kg. In a bag for each substrate. The first layer was filled with the substratum up to 5 cm height. The spawn was sprinkled over it and the bottom of the bags were tied with a rubber to provide a flat circular bottom to the mushroom beds. Similarly, four such layers were filled with the substratum. Inoculation was made with pure grain spawn at 10 gram per kg of substrate on dry weight basis under aseptic conditions. The bags were tied and two vents of one cm diameter were provided.

Spawn running:

After spawning, the beds were incubated in the lab. This is known as spawn running. Spawn run refers to the vegetative propagation of fungal mycelium in the substratum. It was a pre-requisite for the subsequent reproductive growth phase (fruiting).

Incubation: mycelial growth

The spawned compost bags were kept in a dark room until the mycelium has fully penetrated to the bottom of the substrate. In 20 to 30 days, depending upon the substrate/substrate combination, the substrate appears white, due to the growth of the mycelium. The bags were kept for an additional week before they are opened to check that the mycelium is mature enough to fruit. Most strains of the mushroom form primordial after 3 to 4 weeks of mycelial growth. The bags were opened to initiate fruiting, inside a mushroom house. The agro waste Wheat straw, Bajra, Jawar, Corn leaves, corn cob, cotton waste, Rice straw, Bamboo leaves, Sugarcane baggase, were collected from local farms/places and were used for filling the bags. The substrates were chopped to 2-3 cm pieces and soaked in water over night to moisten it and excess water was drained off. After soaking, the substrate was Chemical sterilized. The polythene bags of the size 14 x 22 cm were filled with sterilized substrates and multi layered technique was adopted for spawning. Each bag was filled with 2.5 Kg dry substrate and the spawn was added at the rate of 2 % of the wet weight basis of substrate. Pinning of bags was done for proper aeration. After inoculation the bags were kept in room where the temperature and humidity were maintained around 25 °C and 80 to 90% humidity respectively with sufficient light and ventilation for 20 days. The spawn run was completed within 16 days. The polythene bags were tear-off following the spawn run. Formation of fruit bodies was evident within 3-4 days after removal of poly bags. The beds were maintained up to the harvest of the third flush, which was completed in 35 days after spawning.

Harvesting

As soon as the fruiting bodies developed and attained their full size, they were cut just above the surface of the substrate with sharp knife or blade. Scrape out 1 cm outer layer of the bed after first harvest and did not sprinkle water for 12 hours. From the second day onwards water was sprinkled. Within 3 to 4 days basidiocarps were developed.

Weighing

The fresh weight of basidiocarp at each harvest were taken. The total yield is expressed in terms of bioefficiency being percentage weight of the mushroom on dry weight of substrate.

RESULT AND DISCUSSION:

Different substrates were screened in order to study their impacts on mycelium growth of *Pleurotus eous* and results are given in table 01. From table it is clear that, maximum mycelium growth was recorded due to all substrates at 15 days of incubation period. Moderate to minimum mycelium growth of *Pleurotus eous* was observed at 10 days of incubation period. It is interesting to note that, 10 days of incubation period favoured maximum mycelium growth of *Pleurotus eous* due to bagasse and mixed substrates. On the other hand, mycelium growth was absent in bagasse + wheat flour and bagasse + green gram flour substrate. Bagasse and mix substrate showed moderate growth of mycelium at 5 days of incubation period. Yields of *Pleurotus eous* was observed under the influence of different substrates such as bagasse along with different flours and results are given in table 02. It is revealed from the table that, as compared to control all the substrates gave maximum yield of *Pleurotus eous*. Bagasse + mix flour gave highest yield of *Pleurotus eous* followed by bagasse + wheat flour, bagasse + wheat flour, bagasse + green gram flour. Average yield of *Pleurotus eous* was recorded in case of bagasse + chick pea, bagasse + bajara and bagasse + jowar.

In this study highest yield of *Pleurotus eous* was recorded due to sugarcane bagasse + flour mix. Similar results were observed by Ahmed (1994). He found that, sawdust and sugarcane bagasse were the best substrates for growing of oyster mushroom than other agro-based substrates. Many researchers have used pulse powder for increasing the production of oyster mushroom. Singh (1998) supplemented sugarcane bagasse with chickpea bran at five per cent to produce highest yield of sporophores of *P. abalonus* and *P. florida* followed by bran of pigeon pea, lentil and pea. On the other hand, Dubey (1999) reported that supplementation of substrate (paddy straw) with pigeon pea dal powder at 5% during spawning gave the highest number of sporophores and yielded maximum biological efficiency in *P. sajor-caju*, *P. flabellatus*, *P. ostreatus* and *P. cystidiosus* followed by gram dal and karanj cake. Similarly, (Kathe et al., 1996) found soyabean flour at 3% concentration was found to be better than other nutrients with increased yield of *Pleurotus* spp. significantly. In the next year, Kumar et al., (1997) used pulse powder for spawn production, the study revealed an increased yield of *Pleurotus* spp. supplemented with bengal gram and green gram flour decreased the days required for spawn growth and first harvest and also increased the yield of *P. djamor* and *P. eous*.

Table 1: Effect of different supplements of mycelium growth on *Pleurotus eous*

Sr. no.	Substrate + Flours	Growth			Day of first pin Head
		5 Days	10 Days	15 Days	
1	B + Bajara	+	++	+++	19-01-2020
2	B + Chick pea	+	++	+++	19-01-2020
3	B + Wheat	+	++	+++	19-01-2020
4	B + Jowar	-	+	+++	20-01-2020
5	B + Green gram	-	+	+++	20-01-2020
6	B + Mix	+++	+++	+++	20-01-2020
7	B + Control	+	++	++	20-01-2020

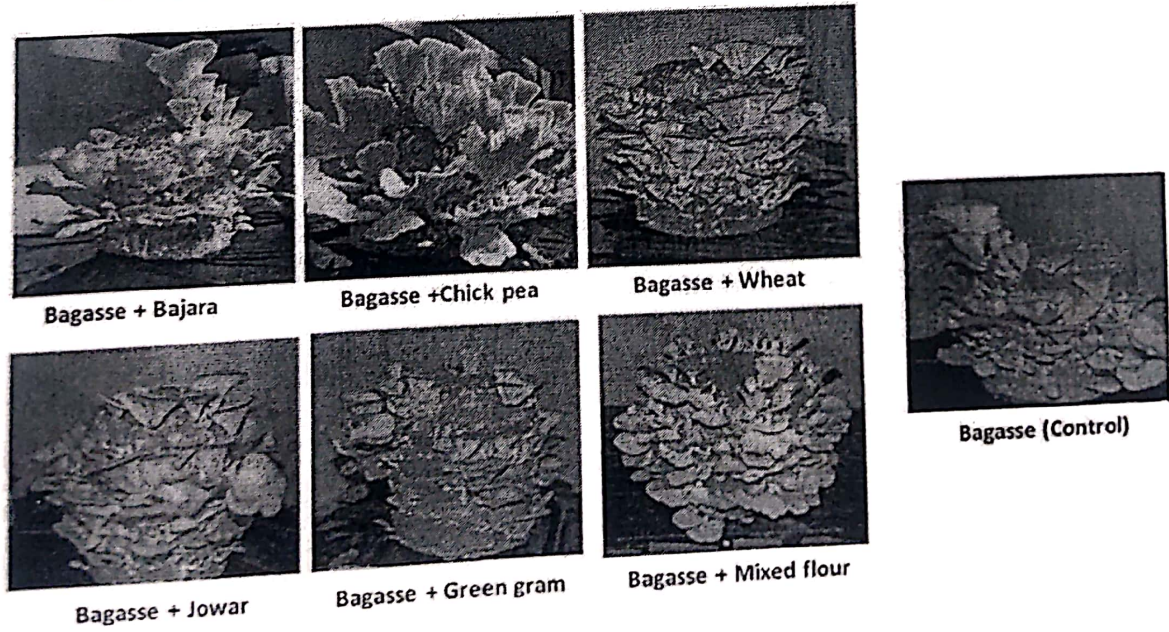
Table 2: Effect of different supplements with baggase on yield on *Pleurotus eous*

Sr. no	Substrate (Bagasse) + Flours	Yield (gm./2 kg of Dry Substrates)			Total Yield (gm)
		I harvesting	II harvesting	III harvesting	
1.	B + Bajara	153.66	172.20	52.10	377.96
2.	B + Chick pea	164.52	180.12	53.17	397.81
3.	B + Wheat	243.10	183.12	65.12	491.92
4.	B + Green Gram	214.50	172.27	45	431.77
5.	B + Jowar	133.30	171.87	38	343.17
6.	B + Mix	316.57	383.20	136	835.77
7.	B + Control	80.24	53.23	36.10	169.57

PHOTOPLATE 1: EFFECTS OF DIFFERENT SUBSTRATES ON MYCELIA GROWTH



PHOTOPLATE 2: GROWTH OF *PLEUROTUS EUOS* ON DIFEERENT SUBSTRATES



CONCLUSION:

From the result it is conclude that, flour of all substrate mixed along with Bagasse was found to be gave maximum yield of *P. eous* B+ wheat and B+Green gram also gave considerable yield of *P. eous*. Hence, It can be said that flour of all the substrate mixed along with Bagasse is found to be good substrate for the growth of *P. eous*. Simultaneously, B+wheat and B+Green gram can be used as far as growth of *P. eous* is concerned. The results of the study stipulate that the performance of *Pleurotus eous* mushroom was highly affected by addition of supplements (flour) into the substrate. The addition of external nutrients increases the productivity of some low-yielding mushroom varieties, and therefore is a useful tool for the industry to introduce new commercially viable varieties. Sugarcane bagasse is a waste material that could feasibly be recycled as a substrate for the growth of mushroom.

REFERANCE

A. Beetz, M. Kustudia, " Mushroom cultivation and marketing," Horticulture production guide (www.attra.ncat.org).2004.

A. A. Kathe, R. H. Balasubramanya and V. G. Khandeparkar, " Cotton stalk spawn of *Pleurotus sajorcaju* and the yield of mushrooms," Mushroom Res. 5: 5-8.1986.

A. Philippoussis, " Production of Mushrooms Using Agro-Industrial Residues as Substrates," In Biotechnology for Agro-Industrial Residues Utilisation: Utilisation of Agro-Residues; Singh nee' Nigam, P., Pandey, A., Eds.; Springer: Dordrecht, The Netherlands; pp. 163– 196.2009.

A. K. Singh. " Cultivation of oyster mushrooms (*Pleurotus* spp.) on sugarcane residues." J.Mycol.Pl.Pathol. 28: 240-245.1998.

B. C. Saha, " Hemicellulose bioconversion. J. Ind. Microbiol. Biotechnol," 30, 279– 291.2003.

Biotech. Letters 10, 369. Mentation of straw by white rot fungi. Eur. J. Appl. Microbiol. Biotechnol., 11: 183-188.

C. Sánchez, " Modern aspects of mushrooms culture technology. Appl Microbiol Biotechnol 64:756– 762.2004.

C. Sanchez , " Cultivation of *Pleurotus ostreatus* and other edible mushrooms," Applied Microbiology and Biotechnology, 85(5): 1321- 1337.2010.

Dawit Abate, " Mushroom cultivation: A practical approach," Berhanena Selam printing press. Addis Ababa. pp. 17-72.1998.

- J. Sun, X. Sun, R.C. and Su, Y. Q. " Fractional extraction and structural characterization of sugar cane bagasse hemicellulose. Carbohydrate Polymers," 56,195-204.2004.
- K. G. Satyanarayana,G.G.C. Arizaga ,and F. Wypych, " Biodegradable composites based on lignocellulosic fibers—An overview," Prog. Polym. Sci. 34, 982– 1021.2009.
- K. S. Visvanathan and N. Gopalaratnam ," Effect of bagasse pulp on quality," A study of various indian paper grades: 1-15. 5.1998.
- K. Mori, T. Toyomasu, H. Nanba and H.Kuroda, " Antitumor Activities of Edible Mushrooms By Oral Administration. Proc. Int' I. Sym. Sci. Tech. Aspect of Culti. Edible Fungi. Penna," State Univ. USA. pp. 49-5 . B., 8, 745-751.1986
- M. A. Ahmed, "Cultivation of Oyster Mushroom," 1st ed. Cairo, Egypt, Comet Co., 63.1994.
- M. H. Morais, A. C. Ramos, N. Matos, E. J. Santos-Oliveira ," Production of shiitake mushroom (*Lentinus edodes*) on lignocellulosic residues: note," Food Sci Technol Int 6:123– 128.2000.
- M. Ritota, and P. Manzi," *Pleurotus* spp. Cultivation on Different Agri-Food By-Products: Example of Biotechnological Application," Sustainability, 11, 5049: 1-26.2019.
- M. Kumar, A. Kumar, H .Kumar and V. K. Singh. " Evaluation of different substrates and supplementation on yield of *Pleurotus sajorcaju* and *P.ostreatus*," In: Indian Mushroom Conference.Poster paper No.2.07, 26. 1997.
- R. Cohen, L. Persky and Y. Hadar, " Biotechnological applications and potential of wood-degrading mushrooms of the genus *Pleurotus*," Appl. Microbiol. Biotechnol. 58, 582– 594.2002.
- R. S. Gupta, " Mushroom Cultivation," Indian Hort., 31(1): 11.1986.
- R. Pathak, N. Joshi, R. R. Dwivedi ," Eco friendly production of *Agaricus bisporus* (*linge*) imbach (white button mushroom)," Net:sdI :6: 57-60.2009.
- S. T. Chang, " Cultivation of *Volvariella* Mushrooms in Southeast Asia," In: Trop. Mushrooms; Biological Nature and Cultivation Methods. The Chinese Univ. Press, Hong Kong. pp. 135-156.1992.
- S. C. Dubey, " Effect of different substrates and amendments on the yield of *Pleurotus* species," J. Mycol. Pl. Pathol. 29:209-213.1999.
- X. Q. Dong, J.S. Yang, J. S. N. Zhu, E.T. Wang, and H.L. Yuan, " Sugarcane bagasse degradation and characterization of three white-rot fungi," Bioresource. Technol. 131, 443– 451.2013.

V. P. Mane, S. S. Patil, A. A. Syed and M. M. V. Bai,. " Bioconversion of low quality ligno-cellulosic agricultural waste into edible protein by *Pleurotus sajor-caju*," (Fr.) Singer. J. Zhejiang Univ. Sci 2007.

Production of shiitake mushroom (*Lentinus edodes*) on ligno-cellulosic residues: note. Food Sci Technol Int 6:123- 12

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