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Comparative Study on Cultivation of Oyster Mushroom *Pleurotus Ostreatus* on Different Substrates

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Abstract:

Mushroom is an edible macro fungus, *Pleurotusostreatus* (oyster mushroom)is an edible mushroom that also has high medicinal values. In this study, the effect of substrate on growth, yield and nutritional composition of domestically-grown oyster mushroom (*Pleurotusostreatus*) was investigated. The substrates used were straw, coir pith, newspaper, sawdust and banana leaves. The substrates were pasteurized with hot water (90°C for 4 h) before spawns of oyster mushroom were inoculated to them. After inoculation, the substrates were kept in a controlled environment. Various chemical characteristics like moisture content, total ash content, protein was estimated. Lastly antimicrobial activity of oyster mushroom was studied using gel diffusion method .The study concluded that *Pleurotusostreatus*(oyster mushroom) can be grown on any of the five substrates, but maximum yield was observed on Banana leaves.

Keywords —*Pleurotusostreatus* ,substrates ,Yield ,Nutrition,antimicrobialactivity,gel diffusion method.

I. INTRODUCTION

Mushrooms are considered to be healthy food that can reduce malnutrition and help the country deliver the global commitment of achieving the Millennium Development Goals (MDG) on health, poverty and hunger (United Nations, 2000). Among many kinds of edible mushrooms, oyster mushrooms of *Pleurotus* spp. has been commercialized and consumed *Pleurotus* is a genus of gilled mushrooms which includes one of the most widely eaten mushrooms, *P. ostreatus*. Oyster mushroom(*pleurotus spp.*) belonging to class Basidiomycetes and family Agaricaceae is popularly known as 'dhingri' in India and grows naturally in the temperature and tropical forests on dead and decaying wooden logs or sometimes on dying

trunks of deciduous or coniferous woods. It may also grow on decaying organic matter.

Pleurotus spp. can make use of the largest variety of waste substrates with its fast mycelial growth and its multilateral enzyme system. A range of about 200 different wastes is available as oyster mushroom substrates (Joseph, 2004). In the early days, mushrooms were grown outdoors in most parts of the world. But, most modernmushroom farmers cannot rely on the natural environment, and hence, build temporarymushroom growing houses and provide good conditions for higher yield of mushrooms. Oyster mushrooms can be cultivated in bags, trays or plastic bottles on lignocellulosic substrates. They grow well on different types of lignocelluloses materials, converting them into digestible and protein-rich substances suitable for

animal feeds. *Pleurotus* spp. may be produced in the tropics on a mixture of sawdust and rice bran, rice straw and rice bran, saw dust and ipil-ipil leaves and other combinations of tropical wastes. Other wastes such as corncobs, cotton waste, sugarcane bagasse and leaves, corn leaves, grasses, rice hulls and water hyacinth leaves are also good substrates for growing this mushroom (Quimio, 1986). The substrates used in each region depend upon the availability of agricultural wastes. A list of substrates used for growing various *Pleurotus* species is shown below:

Substrate	<i>Pleurotus</i> species	Reference
Coir waste	<i>P.sajorcaju</i>	Eyni et.al(1995)
Cotton waste	<i>P.ostreatus</i>	Tan (1981)
	<i>P.sajorcaju</i>	Balakrishnan and Nair (1995)
Hulled maize	<i>P.sajorcaju</i>	Balakrishnan and Nair (1995)
Rice straw	<i>P.florida</i>	Mathews et.al(1996)
	<i>P.sajorcaju</i>	Mathews et.al(1996)
	<i>P.ostreatus</i>	Khandar et.al (1991)
Rubber wood	<i>P.florida</i>	Mathews et.al(1996)
Saw dust	<i>P.sajorcaju</i>	Mathews et.al(1996)
Waste paper	<i>P.sajorcaju</i>	Balakrishnan and Nair (1995)
Wheat straw	<i>P.sajorcaju</i> <i>P.ostreatus</i>	Sharma and Vijay (1996)

The present study focus on the growth of oyster mushroom on different substrates which include Banana leaves(S1), Coir pith (S2), Paper (S3), Sawdust (S4), Straw (S5). Analyse various chemical characteristics of *Pleurotus ostreatus* (oyster mushroom) and Investigate Antimicrobial activity of crude extract of *Pleurotus ostreatus* (oyster mushroom) by gel diffusion method.

II. MATERIALS AND METHODS

Pleurotus ostreatus spawns grown on sorghum grains were collected from Department of Microbiology, Kerala Agricultural University Mannuthy Thrissur Kerala India.

A. Preparation Of Substrates And Mushroom Bed

Different substrate like paddy straw, coir pith, paper, saw dust, banana leaf were taken. The substrate were subjected to soaking for an hour. The substrate were drained off and autoclaved. After sterilization they were shade dried on a clean surface. Substrates were packed in disinfected polypropylene bag and spawn were added correspondingly, 8-12cm inside the margin at an interval of 10-15cm all along the periphery. Later it was pressed gently, and the bags were sealed for spawn development. Small holes were made on the bags for aeration. Spawned bag were hanged in a dark sterile room with the temperature about 30-35°C and humidity was maintained by spraying water twice in a day. Mycelium development was observed. Data were recorded periodically during the growing season namely -1st flush, 2nd flush and 3rd flush. The observation were recorded on the number of primordia, fruiting bodies and fresh weight of mushroom.

B. Determination of Moisture Content

Moisture content of fresh mushroom was determined by weighing mushroom fruits after harvesting, this weight was considered as the initial weight. It was then dried by placing in hot air oven at 105 ± 1°C for 3-5 hours. The dried sample get finely powdered and then weighed constitutes the dry weight.

Moisture percentage can be calculated as:

$$\text{Moisture \%} = \frac{\text{Initial weight} - \text{dried weight}}{\text{Initial weight}} \times 100$$

C. Total Ash Content

One gram of raw mushroom sample was weighed and made into ash by placing in hot air oven, until the sample became a gray ash and it was weighed. The total ash was calculated using the following equation (Raghuramulu et al., 2003).

$$\text{Ash content (g/100g)} = \frac{\text{Weight of ash}}{\text{Weight of mushroom sample taken}} \times 100$$

D. Mushroom Extraction

An extract is a preparation that results from taking raw plant material and applying a solvent to it, such as water or alcohol. The solvent causes the dissolvable compounds to be separated from the structural part of the plant or mushroom. The extraction process breaks down chitin – the indigestible cell wall of fungi that contains the main active ingredients. For these reasons, extracts are frequently used to manufacture dietary supplements. Hot water extraction is the process of using water to dissolve water soluble ingredients of mushroom. In this method, the raw mushroom sample was heated in water at a temperature of 80-170 degree Celsius for a certain period of time till the water evaporated completely. The resultant extract was powdered. Alcohol extraction is the method of using ethanol as solvent to dissolve water insoluble ingredients of mushroom. Ethanol was added to the raw mushroom sample and kept undisturbed in room temperature. It is beneficial to use both water and alcohol extraction – a method referred to as a dual or double extraction. In a dual extract, the material is first extracted with either hot water or alcohol, and then the remaining plant material is extracted again with the other method. The liquid from the first extraction is combined with the liquid from the second extraction, forming a dual extract. Dual extraction is typically used for mushrooms that contain significant levels of both water-soluble and alcohol-soluble active ingredients. The extracted sample was further used as sample for Antimicrobial analysis.

E. Protein Estimation By Biuret method

BSA was chosen as the protein standard and the extracted mushroom absorbance was measured at 550nm. This standard graph was plotted with the concentration of protein on x-axis and the absorbance on y-axis and the amount of protein in the given unknown solution was calculated.

F. Antimicrobial Activity Of Pleurotus Ostreatus By Gel Diffusion Method

The antimicrobial activities of extracts of oyster mushrooms were determined by using Agar well

diffusion method. For this purpose, mushroom extracts and Mueller-Hinton broth were prepared. *E.coli* and staphylococcus Tetracycline (30mcg) and Gentamycin (10mcg) discs were used. After 24 hrs incubation the results were observed

III.OBSERVATION AND RESULTS

Substrate	Colonisation	Pin head formation	Avg. yield	No. of flesh	Cap colour (pin head stage)	Cap colour (mature stage)
Banana leaves (s1)	20 th day	20 th day	160 g	34	Pure white	Pure white
Colr pith (s2)	14 th day	27 th day	10 g	05	Pure white	Pure white
Paper (s3)	17 th day	25 th day	10 g	05	Pure white	Pure white
Saw dust (s4)	14 th day	19 th day	30 g	10	Pure white	Pure white
Straw (s5)	14 th day	19 th day	100 g	15	Pure white	Pure white

Table 1: Average yield of mushroom on different substrate

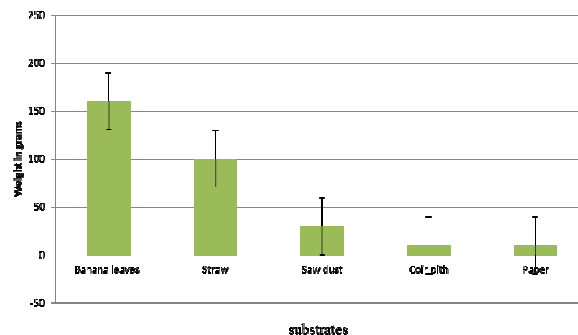
AVERAGE YIELD ON DIFFERENT SUBSTRATES



Figure 1 :Different stages of mushroom growth on substrates

Initial weight	145 g
Dried weight	7.5 g
Moisture percentage	0.0094 %
Ash weight	0.1052 g
Total ash content	10.52 %

Table 2: Moisture and Ash Content of Mushroom

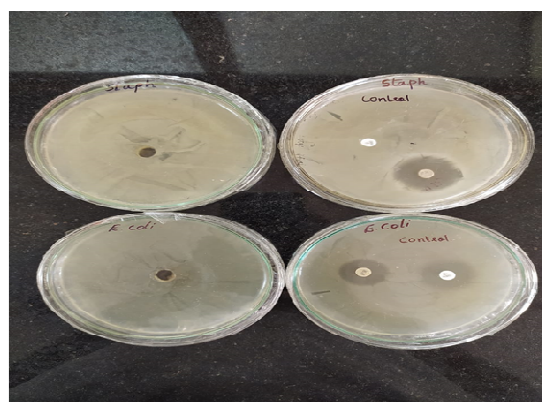
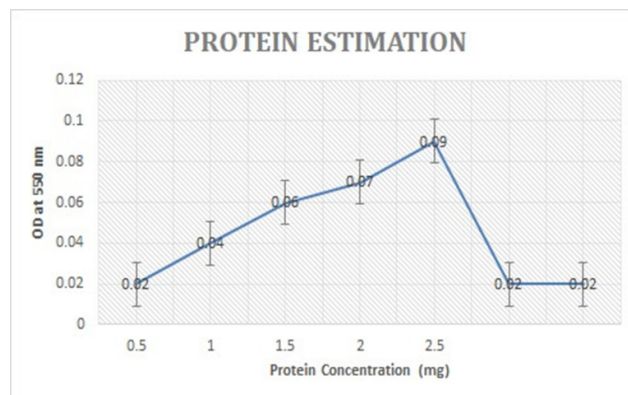


Figure 2:Antimicrobial activity of crude extract of *Pleurotus ostreatus* (oyster mushroom) by gel diffusion method.

IV. CONCLUSION

Oyster mushroom is an edible mushroom which can be grown on various substrates like banana leaves, coir pith, paper, straw, saw dust etc. This study confirmed that the Oyster mushroom, *Pleurotus ostreatus* grown well in banana leaves when compared with other substrates. Total protein content present in fruit

bodies of the oyster mushroom was analyzed by the Biuret method. The protein concentration was finally determined using standard curve with concentration of protein on x-axis and the absorbance on y-axis. And it is found to be 0.02g/ml. In our study of antimicrobial properties of the oyster mushroom, no zone of inhibition was observed, which indicates that both the organism *Escherichia coli* (Gram negative) and *Staphylococcus aureus* (Gram positive) are resistant to the extract.

V. FUTURE PERSPECTIVES.

Pleurotus as health promoter and environmental restorer is gaining more importance as compared to other medicinal mushrooms resulting in an upsurge in their R and D activities during the past two decades. So in near future nutraceutical studies can be focused. The chemical nature of the bioactive compounds present in this mushroom includes: polysaccharides, lipopolysaccharides, proteins, peptides, glycoproteins, nucleosides, triterpenoids, lectins, lipids and their derivatives. The activity of these bioactive compounds can be identified. Studies on spent mushroom can be promoted for zero waste management.

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