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RESEARCH ARTICLE

CULTIVATION OF WILD PLEUROTUS SPECIES ON AGRICULTURAL SUBSTRATES AND EVALUATION OF THEIR NUTRITIONAL PROPERTIES.

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Abstract

Three wild isolates of Pleurotus were collected from different parts of north western Himalayas. Based on the morphological and cultural traits isolates were identified as i.e P.djamore ,P.ostreatus and Pleurotus florida .These three species were cultivated on different locally available substrates in Himachal Pradesh. Different substrates used were paddy straw , wheat straw, soybean straw and maize stalks .All the species of Pleurotus performed well on Paddy straw and soybean straw.. Highest protein content (2.50 mg/g) was found in P. ostreatus on soybean straw followed by Pleurotus florida (2.32 mg/g) on same substrate .

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Introduction:-

Wild edible mushroom have been integrated part of the diet, especially among rural, urban dwellers and tribal people. Some of the common edible mushrooms, which are predominantly consumed in India are Pleurotus sajor-caju, P. florida, P. platypus, P. djamor, Volvariella volvacea and Calocybe indica (Pan et al., 2008; Ramkumar et al., 2010). Pleurotus species is known as oyster mushrooms, which are widely spread saprophytic macrofungi and distributed throughout the temperate and tropical forests of the world (Gunde Cimerman 1999). Oyster mushroom is consumed all over the world due to its taste, flavor, high nutritional value and some medicinal properties. Many species of this genus are rich in proteins with essential amino acids, polysaccharides, essential amino acids, dietary fibers , important minerals and some vitamin (Sharma and Jandaik,2017). Cultivation of oyster mushroom has recently increased tremendously throughout the world because of their abilities to grow at a wide range of agro-based residues. These white-rot fungi are useful decomposers of various agricultural wastes .Pleurotus spp. are the most talented group among the cultivated mushrooms, which have ability to degrade many lignocellulosic substrates and are capable to colonize successfully on these substrates (Patrabansh and Madan, 1997). Agriculture residues are the major source of lignocellulosic materials, which is best substrate for solid state fermentation of edible fungi. Pleurotus species has very high saprophytic colonizing ability and can degrade wheat straw efficiently Zadrzil (1980). Growing oyster mushrooms convert a high percentage of the lignocellulosic materials to fruiting bodies. Therefore, cultivation of Pleurotus species on various agricultural residues offers high value products with nutritional and medicinal properties. Also, mushroom production gives additional or alternative income to farmers looking for a value-added product and a way to supplement farm income while making use of byproducts or co-products from other crops.

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Materials And Methods:-

Collection:-

All the species were collected from the different localities of North West Himalayas

Isolation and maintenance of pure culture:-

Pure culture of collected wild species was raised from the pileus portion of the mushroom where the lamellae join the stipe. For raising the culture a small piece of mushroom tissue was cut and removed with the help of sterilized scalpel under aseptic conditions and subsequently sterilized by dipping in 0.02% mercuric chloride solution and inoculated into the pre-prepared sterilized MEA slants. The inoculated MEA slants were stoppered with the cotton plugs and incubated at $28 \pm 2^\circ\text{C}$ temperature. The total operation of culturing was done aseptically under laminar flow. After 3-4 days (depending upon species), white mycelium started emerging and spreading on the PDA slants. Purification of the culture for further maintenance and utilization in experiments was done through repeated sub-culturing. Then the equal bits of the mycelium were put in petriplates and the growth was measured on daily basis in triplicates.

Spawn Preparation:-

The spawn of selected isolates of *Pleurotus* was prepared following the standard technique.

Evaluation of substrates for sporophore production:-

Pleurotus species were grown on different agricultural wastes like paddy straw, wheat straw, soybean straw and maize stalks. These agricultural wastes were chopped, dried and soaked in water for overnight and then drained off the excess water. The moist substrates were steamed under pressure inside the autoclave for 40 minutes at 15psi. After sterilized substrates were taken out of the autoclave and allowed to cool down at room temperature. Substrates were next day spawned with spawn of different species mentioned above @2.5%. The filled bags were tied with rubber band and 6-8 small holes (1 mm) made around the bags. The mushroom bags were kept in a dark room, where temperature ranged between $24-28^\circ\text{C}$ and relative humidity 80-85% for the spawn run. Polythene bags were teared off after completion of spawn run and removed the side without disturbing the beds of mushroom and kept on wooden racks. To provide adequate moisture, daily water was sprayed on mushroom beds. Usually 3-4 days after opening the bags, mushroom primordial began to form. Fruit bodies were harvested in about 3-4 days after maturity. Harvesting was done by grasping the stalk and gently pulling or twisting the mushroom from substrates level.

Observations:-

were recorded in terms of days to spawn run and first flush, pin head initiation and total yield of fruit bodies.

Nutritional Studies:-

Standard biochemical techniques were employed for determining the proximate composition and nutraceutical components of different species of *Pleurotus* (Fr.) P. Kumn. investigated during the present study.

Estimation of Protein content:-

Protein estimation was done by the method given by Bradford (1976). Estimation of carbohydrates was carried out by following phenol sulphuric acid method (Dubois et al. 1956; Hedge and Hofreiter 1962).

Results:-

Three wild isolates of *Pleurotus* were collected from different regions of North West Himalayas. Collected isolates were identified on the basis of macroscopic and microscopic traits of basidiocarp (Plate 1) and cultural characteristics. Results depicted in table 1 show that Pileus colour of isolate P1 was pink. The surface of pileus was smooth and wavy, stipe length was 1.0-1.5 cm. Spore print was white with spore size of $6.5-9.5 \times 3.0-4.5 \mu\text{m}$. The isolate was identified as *Pleurotus djamora*. Isolate P2 was creamish white in colour surface of pileus was smooth to sticky, spores size was $8.0-9.5 \times 3.0-4.5 \mu\text{m}$. The isolate was identified as *Pleurotus ostreatus*. Pileus colour of isolate P3 was creamish white, surface of pileus was smooth, thick and wavy margins with stipe length of 0.5-1.0cm. Spore print was of white colour. Spore size was $10.3-12.7 \times 5.5-7.8 \mu\text{m}$. Based on macroscopic and microscopic traits the isolates were identified as *P. sajorcaju*. Similarly, many mushrooms were successfully identified based on their morphological and cultural characters by several other workers (Hawksworth, 2004; Fonseca et al., 2008; Gateri, 2008).



Plate1a. P.djamore P.ostreatus P.florida

Table 1:-Morphological characteristics of the Basidiocarp of different Pleurotus species

Isolate	Pileus colour	Pileus Surface	Margins	Stipe length	Spore print	Spore size	Species Identified
P1	Pink	Smooth	Round	1.0-1.5	Creamish	6.5-9.5 X 3.0-4.5	P.djamore
P2	Cremish white	Smooth /sticky	curved	0.5-1.0	White	8.0-9.5 X 3.0- 4.5	P.ostreatus
P3	Creamish white	Smooth, thick	wavy	0.5-1.5	White	10.3-12.7 x 5.5-7.8	P.florida

The results depicted in table 2 reveals that maximum yield was obtained on paddy straw (standard substrate) followed by soybean straw irrespective of the species. Among different species it was found that P. ostreatus resulted in maximum yield (840 g/kg) followed by P. florida (828.80 g/kg). All the three species resulted in minimum yield on maize stalks. Pleurotus species are known to grow relatively well on cellulosic /carbohydrate materials as reported by other researchers Zadrazil (1978) and Krishnamoorthy (1981).

Table 2:-Comparative evaluation of different substrates on yield of different Pleurotus species

Pleurotus species	Substrates	Mushroom growth period in days after spawning		
		Spawn run period g/kg	Pinhead initiation	Total Yield (g/kg)
P.djamore	PS	15.00	19.00	798.00
	SS	17.66	23.00	771.20
	MS	19.00	21.00	463.86
	WS	21.00	22.00	547.86
P.ostreatus	PS	15.33	19.00	840.00
	SS	20.00	23.00	749.16
	MS	20.66	25.00	415.83
	WS	20.33	24.00	525.83
P.florida	PS	16.00	20.00	828.80
	SS	21.33	22.00	802.00
	MS	18.66	25.00	453.60
	WS	20.66	24.00	518.00
C.D.(1%)		2.36	2.68	47.9

Paddy straw =PS, Soybean Straw =SS, Maize stalks= MS, Wheat straw=WS

Table 3:-Estimation of Proteins and Carbohydrates content of three Pleurotus species on different substrates

Substrates	Pleurotus djamore		Pleurotus ostreatus		Pleurotus florida	
	Total Protein (mg/g)	Total Carbohydrate (mg/g)	Total Protein (mg/g)	Total Carbohydrate (mg/g)	Total Protein (mg/g)	Total Carbohydrate (mg/g)
PS	2.00	3.81	1.95	2.91	2.45	2.95
SS	2.10	2.74	2.32	2.09	2.50	2.82
MS	1.84	3.07	1.94	3.51	1.94	3.11

WS	2.09	3.15	2.15	3.60	2.10	3.83
CD	0.64	0.18	0.26	0.13	0.72	0.12

Paddy straw =PS, Soybean Straw =SS, Maize stalks= MS, Wheat straw=WS

Relatively higher amount of the protein content (2.50 mg/g and 2.32 mg/ gm) was recorded in *Pleurotus ostreatus* and *Pleurotus florida* respectively grown on soybean straw. The lowest protein content (1.84 mg/g) was recorded in *P. djamore* grown on maize stalks. *Pleurotus florida* showed highest carbohydrate content (3.83 mg/g) content on wheat straw. Total carbohydrate content ranged from 2.09 mg/g to 3.83 mg/g in all the *Pleurotus* species on different substrates. The digestibility of edible mushroom *Pleurotus* species proteins is as same as that of plants (90%) and comparable with meat (99%) (Wani et al., 2010). However, the average of protein content of *Pleurotus* is most common between 20 and 25% (Patil et al., 2010). Syed et al., (2009) cultivated *P. florida* on different agro-wastes and reported that soybean straw showed significantly highest yield with maximum crude protein of 23.50 % on dry weight basis.

Naraian and Dixit reported maximum Protein and Carbohydrate content in *P. sajor-caju* and *P. florida* grown on wheat straw.

Conclusions:-

The result of present studies revealed that *Pleurotus* species can be used for agricultural waste management food supplements to serve as cheap source of protein to solve protein deficiency in poor people. The study also revealed that morphological characteristics and nutritional composition of mushroom depend on the substrate used for cultivation.

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