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Isolation, porification & maintenance of pure culture and growth of oyster mushroom (*p. sajor caju)* on difference solid media

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Abstract
Experiments were conducted at Department of Plant Pathology, T.D.P.G. College, Jaunpur in the two consecutive year 2010-2011 and 2011-12.Edible mushroom *Pleurotus sajor caju (Fr.)*' Singer to evaluate the Growth of *P. sajor caju* on difference solid media.The experiments were laid out in completely Randomized design. The pure culture of *P. Sajor caju* obtained from mushroom research lab department of Plant Pathology, N.D.U.A.T., Faizabad. Isolation & Porification done by standard method. Growth of *P. Sajor caju* tested on different non-synthetic & synthetic solid media. To find out the best suited medium for growth of the *P. sajor caju* under present study, The experiment consisted of two type of solid media viz. (a) Non-Synthetic (Potato dextrose agar medium, Oat meal Agar Medium and Corn Meal Agar Medium), (b) Synthetic (Richard's Medium, Czapek's (Dox) agar Medium and Brow's Starch Agar Medium). The different constituents of media were dissolved in distilled water separately and for solidification 2% agar was added. In case of Potato dextrose, Oat meal, cornmeal extraction were taken after boiling them on a water bath. All the media were sterilized in the autoclave at 1.1 kg pressure/cm^2 for 20 minutes. The petridishes were handled aseptically in a inoculation chamber using a sprit lamp flame. The medium in the plates was allowed to solidify before inoculation with *P. Sajor caju*. During both the years solid media of Non-Synthetic, Potato Dextrose Agar medium were observed significantly higher than other tested, medium. Czapek agar medium was also significantly superior than oat meal (7.4 cm of 7 days after inoculation). Lowest mycelial growth was observed on Brow's agar medium.

Keywords: Non-Synthetic, Agar Medium, Inoculation and Potato Dextrose

Introduction
The history of the man is the record of hungry creature in search of food on this planet and the use of edible fungi amongst his menus is a civilization itself. Mushrooms have been part of fungal diversity for around 300 million years ago. The word Mushroom been derived from the French "mousseron", "mousse" or moss known by several names viz. puffballs, morels and truftles (Ramsbottom, 1953). In India, mushrooms vernacularly known as 'Khumbhi', 'Chhatra', Kukurmutta', 'Dhingri', Dharti Ka Phool etc. Mushrooms are the member of higher fungi, which lack of chlorophyll i.e., they can't utilize solar energy to manufacture their own food as green plants. However, mushroom can produce a wide range of enzymes that degrade, the complex substate on which they grow, following which they absorb the soluble substrate for their own nutrition. The term mushroom is broadly defined as a macro fungus with a distinctive fruiting body which can be either epigeous or hypogeous and large enough to be seen with the naked eye and to be picked by hand (Chang and Miles, 1993) [4]. Taxonomically mushrooms belong to phylum Basideomycotina and Ascomycotina (Alexopolous *et al.*, 1996) [1]. They occur naturally and seasonally in various habitats and nicks all over the world. The Mushrooms comprise a large heterogeneous group having various shapes, sizes, colour, all quite different in character, appearance and edibility. Out of this large group, with more than 2000 edible species, about 300 species belonging to 70 genera are reported from India. However, only a few have been brought under cultivation on commercial scale. Out of 2000 species of prime edible mushrooms, about 80 have been grown experimentally, 20 cultuvated commercially and six namely *Agricus bisporus, Lentinula edodes, Pleurotus, Auricubria, Valvarella* and *Flammulina* produced on industrial scales (Chang and Miles, 1991) [5] and thus

Called as big six mushrooms, of these, the *Pluerotus* mushrooms is generally referred to as 'Oyster mushroom' all over the world. In India it is commonly known as Dhingri mushroom. It has gained importance only in the last decade and is now cultivated in many countries in the subtropical and temperate zones. *Pleurotus* has gained its name from the Greek world "Pleuro" which means formed laterally or in a side way position, referring to the lateral position of the stipe in relation to pileus (Jandaik 1997) [8]. The world 'Osyster' refers to the oyster like apperance of the fruit bodies. According to the systemic position, Pleurotus belongs to the Phylum Basidiomycota, class basidiomycetes, order Agraricales and family Pleurotaceae (Tricolomataceae), (Kirk *et al.,* 2001) [9]. In the nature species of *Pleurotus* are generally found as saprophytes, growing on dead and decaying part of wood. However, the first attempt to grow this fungus for human consumption was made by Falck (1971) [6] in Germany. The process of cultivation on readily available subtrates was taken up earlier in India on paddy straw (Bano and Srivastava, 1962) [3] and in Japan on saw dust (Schanel *et al.* 1966) [13]. This attracted attention of a number of researchers all over the world and today various species of *Pleurotus* are appreciated for their culinary attributes and broad adoptability under varied agro-climatic conditions. China has become an enormous producer and consumer of cultivated edible mushrooms and also a major producer of medicinal mushrooms. Total production of China in 1997 was 3.910 million tonnes which amount to 63.6% of the total world output. However, in India, mushroom production started in 1960's. In the year 1985 total mushroom production was only 4000 tones, which reached to 30,000 tones in 1995. Further it was increased to 70000 tones in 2004. The state involved in mushrooms production in India or Tamiladu, Maharastra, Punjab, Harayana, Uttaranchal, U.P. and Andhra Pradesh. Upadhyay and Vijay (1991) [15] tried five selected species of *Pleurotus* i.e. *P. ostreatus, P. florida, P. fossulatus, P. eryngii and P. corrnucopiae* for yield. Highest yield of 94 percent biologgical efficiency was obtained with *P. florida* followed by 32 percent biological efficiency for *P. ostreatus* on wheat straw at low temperature ranging 12-16^{0}C and relative humidity 65-75 percent.

Material and Methods

Experiments were conducted at Department of Plant Pathology, T.D.P.G. College, Jaunpur in the two consecutive year 2010-2011 and 2011-12.Edible mushroom *Pleurotus sajor caju (Fr.)*' Singer to evaluate the Growth of *P. sajor caju* on difference solid media.The experiments were laid out in completely Randomized design. One hundred litres of tap water was filled in a plastic drum of 200 litre capacity. A stock solution with 125 ml formaldehyde and 7 g Bavistin in water. This solution was stirred properly with a slick for its mixing. Now 10 kg dry straw abstrate was steeped completely in this chemical solution. The mouth of the container was closed with the lid and kept as such for 18 hours. After 18 hours the straw was taken out from the chemical solution and put on a wire sieve for removal of extra solution. It was then spread in thin layers over a clean coneneted floor for further removal of excess moisture.

The pure culture of *P. Sajor caju* obtained from mushroom research lab department of Plant Pathology, N.D.U.A.T., Faizabad. Isolation & Porification done by standard method. Growth of *P. Sajor caju* tested on different non-synthetic & synthetic solid media. To find out the best suited medium for growth of the *P. sajor caju* under present study, The experiment consisted of two type of solid media viz. (a) Non-Synthetic (Potato dextrose agar medium, Oat meal Agar Medium and Corn Meal Agar Medium), (b) Synthetic (Richard's Medium, Czapek's (Dox) agar Medium and Brow's Starch Agar Medium). The following six different non-synthetic & synthetic solid media were used.

(A) Non-Synthetic

(i) Potato dextrose agar medium

Peeled Potato	-	200.00 g
Agar	-	20.00 g
Dextrose	-	20.00 g
Distilled water	-	1000 ml

(ii) Oat meal Agar Medium

Oat meal	-	50.00 g
agar	-	20.00 g
Distilled water	-	1000 ml

(iii) Corn Meal Agar Medium

Corn Meal	-	50.00 g
Agar	-	20.00 g
Distilled water	-	1000 ml

(B) Synthetic

(iv) Richard's Medium

Potassium nitrate (KNO_3)	-	10.00 mg
Potassium dihydrogen Phosphate (KH_2PO_4)	-	5.00 g
Magnesium sulfate ($MgSO_4$-H_2O)	-	2.50g
Ferric chloride ($FeCl_2$)	-	0.02 g
Sucrose	-	50.00 g
Agar	-	20.00
Distilled water	-	1000 ml

(v) Czapek's (Dox) agar Medium–

Magnesium Sulfate ($MgSO_4$-H_2O)	-	0.50g
Potassium dihydrogen Phosphate (KH_2PO_4)	-	1.00 g
Potassium Chloride (KCL)	-	0.50 g
Ferrous Sulphate ($FeSO_4$)	-	0.01 g
Sodium Nitrate	-	2.00 g
Sucrose	-	30.00 g
Agar	-	20.00
Distilled water	-	1000 ml

(vi) Brow's Starch Agar Medium–

Potassium Phosphate (KPO_4)	-	1.36 g
Sodium carbonate ($NaCO_3$)	-	1.06 g
Magnesium Sulphate ($MgSO_4$)	-	5.00 g
Dextrose	-	5.00 g
Asparagins	-	1.00 g
Agar	-	1.00 g
Distilled water	-	1000 ml

Preparation of Media

The media were prepared by standard method as described by Riker and Rikber (1936). The different constituents of media were dissolved in distilled water separately and for solidification 2% agar was added. In case of Potato dextrose, Oat meal, cornmeal extraction were taken after boiling them on a water bath. All the media were sterilized in the autoclave at 1.1 kg pressure/cm^2 for 20 minutes. The glasswares were cleaned with chromic acid and distilled water was used during

the entire study relating to media study. Glasswares sterilized at 160^0C for two hour in hot air oven.

Pouring of media in Petriplates

Previously sterilized petridishes used for pouring the medium. A set of petridishes was maintained for each treatment and 15 ml sterilized, melted but cooled medium (about 45^0C) was aseptically poured in each petridishes. The petridishes were handled aseptically in a inoculation chamber using a sprit lamp flame. The medium in the plates was allowed to solidify before inoculation with *P. Sajor caju*.

Inoculum

The inoculum of *P. sajor caju* was grown PDA for 5 days at 25^0C. Small discs (5mm in diameter) were transferred to Petridishes containing the test medium. A set of 5 petridishes was used for each treatment. Linear growth of measured in mm and recorded when the mycelium reached the edges of the Petridishes.

Results and Discussion

It is apparent from Table 1 that the results with various different non-synthetic & synthetic solid media Data show that, PDA Media was the best tested media of fungal growth. After seven days inoculation fungal growth rate was 5.30cm/day followed by czopek agar fungal growth rate was 5.04cm/day. The other medium of fungal growth rate richards agar was 4.18cm/day. The medium of corn meal agar fungal growth rate was 4.10cm/day. The oat meal agar fungal growth rate was 4.07cm/day. The lowest fungal growth rate was 2.71 of brow's agar.

Potato Dextrose Agar medium was significantly superior than other tested, medium. Czapek agar medium was also significantly superior than oat meal (7.4 cm of 7 days after inoculation). Lowest mycelial growth was observed on Brow's agar medium. Sisto *et al.*, (1998) [14] cultivated *Pleurotus* mushroom on new media by mixing traditional constituents. Such as wheat straw and dried root beet residues with residues of tomato industrial processing, grapevine pruning, olive waste water, dregs of pressed grapes, triturated carobs and hay from a palyphytic pasture field, compared with traditional compost in controlled environment conditions and glass containers. *P. eryngii* grew and produced well shaped basidiocarp on all tested substrate.

The pure culture of *P. sajor caju* obtained from mushroom research lab, N.D.U.A.T., Faizabad. Pure culture of *P. sajor caju* tested on different synthetic and non-synthetic media. Potato Dextrose Agar Media, was the best tested media of fungal growth. Average rate of fungal growth was also high on Czapek agar media than Richard's agar, Corn meal, oat meal, and Brow's agar). Gibriel, A.Y. *et.al.* (1996) [7] had also reported Potato Dextrose Agar media extract as liquid or solid media was the best medium tested for both rate and amount of fungal growth. Kligman (1943) [10], Sohi *et al.* (1986) [15] and Sawashe and Sawant (2005) [12] have also made some similar observations.

Table 1: Effect of different media on the growth rate of *P. sajor caju*

Days after inoculation	Medium					
	Potato Dextrose	Corn Meal	Oat Meal	Richards agar	Czopek Agar	Brow's Agar
1	1.0	1.2	1.0	1.2	0.9	0.9
2	2.7	2.0	2.1	2.4	2.6	1.8
3	4.2	3.4	2.8	3.6	3.8	2.4
4	5.6	4.3	3.9	4.3	5.1	3.0
5	7.1	5.5	5.0	4.8	6.3	3.4
6	8.1	5.8	6.3	5.6	8.2	3.7
7	8.6	6.8	7.4	7.4	8.4	3.8
R	5.30	4.10	4.07	4.18	5.04	2.71

R = Average of Mycelial Growth Rate (cm/day)

References

1. Alexopolous CJ, Mims CW, Blackwell M. Introductory Mycology, John Wiley and Sons INC, 1996, 869.
2. Anonymous. Annual Report. National Research Centre for Mushroom, Solan (HP.) India, 1998-99, 30-32,
3. Bano Z, Srivastava HC. Studies on cultivation of *Pleurotus* spp. on paddy straw. Food Sci. 1962; 11:363-365.
4. Chang ST, Miles PG. Mushroom biology a new discipline. *The Mycologist*, (UK), 1993.
5. Chang ST, Miles PG. Recent trends in world production of cultivated edible mushrooms. Mushroom. J. 1991; 504:15-18.
6. Falck *R.* Uber die walkulter des Austernpilzes *(Agaricus ostreatus)* auf laubholzstubben z. *Forst. Jogdwes.* 1917; 49:159-165.
7. Gibriel AY, Ahmed M, Rasmy N, Rizk I, Abdel Rehem, NS. Cultivation of Oyster Mushrooms (*Pleurotus* spp.): Evaluations of different media and organic substrates. Mushroom Biology and Mushroom products, Royse (ed.) ISBN 1-883956-01-3, 1996.
8. Jandaik CL. History and development of *Pleurotus* cultivation in the world and future prospects. In.: Advances in Mushroom Biology and Production. Rai, R.D., Dhar, B.L. and Verma, R.N. (eds.) Mushroom Society of India, NRCM, Cham"baghat, Solan, 1997, 181-192.
9. Kirk PM, Cannon PF, David JC, Stalpers JA. Adnsworth and Bisby's Dictionary of Fungi. Biddies, London, 2001, 655.
10. Kligman AM. Some culture and genetic problems in the cultivation of the mushroom Agaricus campestris. American J Bot. 1943; 30:745-763.
11. Ragunathan R, Gurusarny R, Palaniswamy M, Swaminathan K. Cultivation of *Pleurotus spp.* on various agro-residues. Food Chem. 1996; 55(2):139-344.
12. Sawashe SG, Sawant DM. A study on monoculture isolation and interspecific hybridization of Pleurotus. J Maharashtra. Agri. Urdu. 2005; 30(1):49-59.
13. Schanel LI, Herzig M, Dvorak Z. Veznik: Zpusob vyuziti mene hodoaotynch. Druhu slamy ke Krmym ucelum. Ziepsovaci Navrh C. 13: 14 (c.f. Advances in Mushroom Biology and Production) (R.D. Rai; B.L. Dhar and Verma, R.N. eds.) NRCM, Chambaghat, Solan, 1966.

14. Sisto D, Faggiano S, Mannerucci F. New growing media for cultivation of *Pleurotus* mushrooms. Agricoltura Recerca (Italy). 1998; 20(176):47-50.
15. Sohi HS, Upadhyay RC. Effect of temperature on mycelial growth of *Pleurotus* spp. and their yield performance on selected substrate. Mush. Sci. 1985; 2(1):49-56.