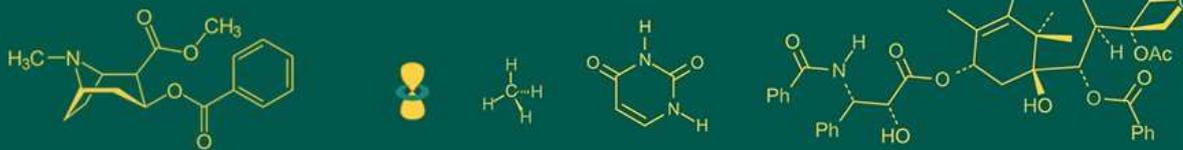


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Effect of different in-organic additives on growth of spawn and production of *Pleurotus* species (*P. djamor* and *P. sajor-caju*)

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Abstract

The present study is growth of spawn and yield of *Pleurotus* spp. against different inorganic additives in laboratory and crop room. Three different type inorganic additives viz. ferrous sulphate, zinc sulphate and magnesium sulphate @ 0.5 and 1.0% were mixed as additive with wheat grain in three replications. Maximum spawn growth of *P. djamor* (90.00 mm) was recorded in Magnesium sulphate 0.5% and zinc sulphate 0.5%. While in case of *P. sajor-caju*, maximum spawn growth (90.00 mm) was recorded in ferrous sulphate 0.5% and zinc sulphate 0.5%. The maximum growth rate of *P. djamor* (6.00 mm/day) was recorded in zinc sulphate 0.5% and magnesium sulphate 0.5%. While in *P. sajor-caju* maximum growth rate (6.00 mm/day) was recorded in zinc sulphate 0.5% and ferrous sulphate 0.5%. The maximum yield of *P. djamor* was observed in ferrous sulphate 1.0% (462.66 gm/kg of dry substrates with 46.26% biological efficiency). In case of *P. sajor-caju* maximum yield was observed in ferrous sulphate 1.0% (790.33 gm/kg of dry substrates with 79.03% biological efficiency).

Keywords: Inorganic, additives, spawn and *Pleurotus*

Introduction

Mushroom farming in India is becoming successful and also popularized day by day because of its very low input, which can bring a significant change in rural economy. The climatic conditions of the region have been found to be ideal for such an attempt. Research and field experiments on production and marketing of several varieties of mushrooms have proved its significant potentiality as a major source of income for rural people (Shahi, V. *et al.*, 2018) [10].

To make Oyster mushroom cultivation more profitable and popular, different types of agro wastes, crop residues and by-products can be used with cylindrical block system, which has already been proven economically viable rather than conventional polybag method (Maniruzzaman 2004) [12]. Oyster mushroom is the third grown mushroom in the world and ranks second in India. For the successful cultivation of oyster mushroom on a small scale or commercial scale, one of the most important requirements is the mushroom seed (Spawn). This is a pure culture of the mycelium grown on a special medium. The production of spawn is done by professionals in the laboratory under controlled conditions or temperature, light and humidity. The contents of Na, K, P, Ca, Mg and Fe for each of the oven dried, vacuum dried and sun-dried mushroom powders. The results of minerals were for Na content 12.41, 9.76 and 15.32 mg/100 gm, for K content 299.5, 226.5 and 295.1 mg/100 gm, for P content 9.19, 762 and 974 mg/100 gm, for Ca content 34.1, 55.5 mg/100 gm and 98.0, for Mg content 297, 254, and 297 mg/100 gm, for Fe content 5.24, 5.78, and 11.6, 297 mg/100 gm for each of the oven dried, vacuum dried and sun-dried mushroom powders, respectively (Maray, *et al.*, 2017) [7]. It can be concluded that the highest values of minerals content in the case of sun-drying followed by oven drying and the lowest values of minerals content in the case of vacuum dehydration (Mattila *et al.*, 2001) [8]. Oyster production is a meager 1200 tonnes production during 1985, the present time total production of oyster mushroom in India reached 21272 metric tonnes in 2016 (DMR, Solan 2016-17). The cultivation of oyster mushroom is now becoming popular in developing countries amongst growers due to

easy and cheap cultivation technology, high nutritive value, good shelf life, agreeable distinctive taste, abundant availability of agro-wastes, choice of species and suitability to varying agro-climatic conditions (Rajaratnam *et al.*, 1989) [9].

Different *Pleurotus* species are reported to show much diversity in their adaptation to the varying agro-climatic conditions and yield potential depending on the substrates nutrient quality. Keeping above importance in view, four chemicals in different doses have been tried to increase the productivity of two species of oyster mushroom namely *P. djamor* and *P. sajor-caju* for growth, spawn run, number of fruiting bodies and yield Gorai, B and Sharma, R. (2017) [2].

Materials and Methods

The present study is growth of spawn and yield of *Pleurotus* spp. against different inorganic additives in laboratory and crop room. The study were conducted during 2018 at Mushroom Laboratory Department Plant Pathology, S. V. P. University of Agriculture and Technology, Meerut, UP, India, which is situated on the Western side of the Delhi-Dehradun high way (NH-58) at a distance of 10.0 km away in the north of Meerut city. The district Meerut is situated between 29°01'N latitude and 77° 45'E longitude at an altitude of 237 meters above the mean sea level.

Establishment of pure culture

Culture of *Pleurotus* spp. were purified and maintained by single hyphal tip method. For this purpose, the culture was grown in sterilized Petri plates on Potato Dextrose Agar Medium (PDA) for 8-10 days. Single branched hyphae from the periphery of the growing colony were marked under low power (10X) in the compound microscope and transferred to PDA slants. These tubes were incubated at 21-24 °C for about a week, again sub cultured on PDA and then stored in a refrigerator at 5-10 °C for further use.

Spawn Production

Three different type inorganic additives *viz.* ferrous sulphate, zinc sulphate and magnesium sulphate @ 0.5 and 1.0% were mixed as additive with wheat grain. For this study, the spawn was prepared in half litre capacity wide mouthed glass bottles. The grains were cleaned to remove any broken, shrivelled grains either by sieving or winnowing or by hand picking of undesired grains. After this, the grains were soaked overnight in clean water and then washed. They were boiled in water for 15 minutes taking care that grains should not split but remain slightly hard after boiling. The boiled grains were spread in thin layer over a wire net to remove excessive water and enable them to cool about 25- 30 °C. The cooled grains were then mixed with 1.2 percent commercial grade gypsum (CaSO₄) and 0.3 percent calcium carbonate (CaCO₃). Gypsum prevents the sticking of wheat grains together and calcium carbonate maintains the pH 5.5-7.5 (Jain, 2005) [4]. The grains were filled up to (100 mm) in the bottle in three replicates. The bottles were plugged with nonabsorbent cotton and covered with butter paper. These bottles were then sterilized at 121 °C (15 lbs pressure) for 2 hours on two consecutive days. Sterilized bottles were taken out from the autoclave, while still hot and were shaken to avoid clumping of grains. Sterilized bottles were inoculated by 9 mm disc in individual bottle. The spawn bottles were incubated without shaking at 24±1 °C in BOD incubator and observations were

recorded on 5th, 10th and 15th day till to completely cover by mycelial growth in bottles (Stamets, 2000) [11]. The experiment was laid out in Completely Randomized Design (CRD) with three replicates of each was applied and the data was obtained were analyzed statistically. Analysis of variance technique and critical difference was calculated at five percent level of significance for comparison with other treatment.

Substrate Preparation

Wheat straw was used as substrate for this experiment. It was soaked (10 kg wheat straw/100 liter water) in a tank with solution of Carbendazim (8 gm/100 liter water) + Formalin (120 ml/100 liter water) for 18 hr (Tank should be covered with polythene sheet to prevent the evaporation of formalin) Thereafter, straw was taken out from the solution and kept for 2-3 hours to drain out the excess water.

Spawning

Spawning was done under aseptic condition. Different sugars spawn of *Pleurotus* spp. (Preparation method described under spawn production) was mixed in Wheat straw (Substrate) @ 4 percent per kg on dry weight basis and 3 kg substrate (containing 60-75% moisture) filled in each polythene bags (22×12") in three replications and made 8-10 holes in each bags for aeration. After spawning bags were kept in the spawn running room under dark condition.

Spawn run

In crop room temperature (22 to 26 °C) and relative humidity (80 to 90 percent) was maintained during spawn run. Humidity was maintained by water spraying three times a day. After the compilations of spawn run in the straw it becomes a compact mass which also sticking to the polythene bags and bags polythene were cut and opened for sporophores formation kept in cropping room. At the time of sporophores formation, the windows were kept open for 1–2 hour to provide fresh air, to release CO₂ and to maintain the relative humidity at 80-90 per cent inside the crop room. Total cropping period given was about 60 days.

Sporophores production

After spawn run, compact stack of substrate (wheat straw) were kept in crop room for the sporophores production. The fruiting bodies were started to appear in 6-8 days. The sporophores were harvested 3-4 days after pinhead initiation. These were harvested by one gentle twisting at the base, taking care that the broken stumps were not left there to avoid rotting in the remaining flushes of running crop. 3-4 flushes were taken after that very few fruiting bodies appear. After the first two flushes, the spawn run blocks were over turned to allow the lower surface and the base to produce fruiting bodies. A total time for cropping up to 3rd flush is about 60-70 days. Watering of the crop is quite important which must be done with a mist sprayer. The water spraying should be done by sprinkler on the blocks after the fruit body start coming up but the floor and walls of the mushroom crop room must be kept moist to maintain requisite humidity (80-90 per cent). Adequate ventilation in the crop room was provided by opening the doors and windows at night for a short time. The fruiting bodies must be protected from direct sunlight but some diffused light (2500-3000 Lux) should be allowed to induce fruiting body formation.

The crop room floor and wall were sprayed with 0.1 per cent Malathion or Sevin and/or light trap to protect it from insect infestation. To prevent the fungal infection, two sprays of Carbendazim 0.02 per cent were given. The observations were recorded as total yield (gm/kg dry straw) and minimum days for spawn run (DFSR), days for pinhead formation (DFPF), days for first harvesting (DFFH), number of fruiting body per beg (NOFB), maximum average weight of fruiting body (gm/FB) and Biological efficiency (B.E.%).

Result and Discussion

Effect of different inorganic on spawn production:

This experiment was conducted to study the effect of different inorganic supplements on spawn growth of *P. djamor* and *P. sajor-caju*. The observations were recorded on 5th, 10th, and 15th days, respectively as shown in Table-1.

On 5th day in case of *P. djamor*, maximum spawn growth (36.33 mm) was recorded in Zinc sulphate 0.5% followed by zinc sulphate 1% (36.00 mm) and minimum spawn growth (28.00 mm) was recorded in control. While in case of *P. sajor-caju*, maximum spawn growth (37.66 mm) was recorded in ferrous sulphate 0.5% and zinc sulphate 0.05%. However minimum spawn growth (34.66 mm) was recorded in control.

On 10th day in case of *P. djamor*, maximum spawn growth (56.33 mm) was recorded in Magnesium sulphate 0.5% followed by zinc sulphate 0.5% and zinc sulphate 1% (56.00 mm) and minimum spawn growth (48.33 mm) was recorded in control. While in case of *P. sajor-caju*, maximum spawn growth (57.33 mm) was recorded in ferrous sulphate 0.5% followed by zinc sulphate 0.5% (56.66 mm). However minimum spawn growth (49.66 mm) was recorded in control.

On 15th day in case of *P. djamor*, maximum spawn growth (90.00 mm) was recorded in Magnesium sulphate 0.5% and zinc sulphate 0.5% which was followed zinc sulphate 1% (87.66 mm) and minimum spawn growth (75.00 mm) was recorded in control. While in case of *P. sajor-caju*, maximum spawn growth (90.00 mm) was recorded in ferrous sulphate 0.5% and zinc sulphate 0.5%. However minimum spawn growth (67.66 mm) was recorded in control.

In case of *P. djamor* maximum growth rate (6.00 mm/day) was recorded in zinc sulphate 0.5% and magnesium sulphate 0.5% and minimum growth rate (5.00 mm/day) was recorded in control. While in *P. sajor-caju* maximum growth rate (6.00 mm/day) was recorded in zinc sulphate 0.5% and ferrous sulphate 0.5%. However minimum growth rate (4.51 mm/day) was recorded in control.

The results were in accordance with the findings of Kumar *et al.*, (2011) [6] reported eight chemicals *viz.* salicylic acid (0.1%), ferrous sulphate (1%), copper sulphate (1%), manganese sulphate (1%), potassium sulphate (1%), magnesium sulphate (1%), zinc sulphate (1%) and calcium carbonate (1%) were tried to see their influence on five species of oyster mushroom (*P. sajor-caju*, *P. florida*, *P. flabellatus*, *P. fossulatus* and *P. sapidus*) with respect to radial growth. Maximum radial growth was observed at 6th days in ferrous sulphate and copper sulphate supplemented medium in *P. sajor-caju* (8.56 & 9.00 cm), *P. florida* (9.00 & 8.75 cm), *P. flabellatus* (8.26 & 8.50 cm), *P. fossulatus* (8.45 and 8.93 cm) and *P. sapidus* (9.00 and 8.50 cm), respectively.

Effect of different inorganic additive on yield of *Pleurotus* spp.

The experiment was conducted to find out the effect of different inorganic additive *viz.* ferrous sulphate 1.0% & 1.2%, copper sulphate 1.0% & 1.0 and zinc sulphate 1.0% @ 1.2 on yield of oyster mushroom (*P. djamor* and *P. sajor-caju*) observation on mycelial ramification in wheat substrates, days for spawn run, days for pin head formation, days for first harvesting, number of fruiting bodies and total yield were recorded as shown in Table 2.

In case of *P. djamor* maximum yield was observed in ferrous sulphate 1.0% (462.66 gm/kg of dry substrates with 46.26% biological efficiency) which was followed by ferrous sulphate 1.2% (456.66 gm/kg of dry substrates with 45.66% biological efficiency) while minimum yield was observed in control (409 gm/kg dry substrates with 40.90% biological efficiency) which was followed by zinc sulphate 1.2%, (418.33 gm/kg of dry substrates with 41.83% biological efficiency) which was statistically lower than all treatment. Minimum days for spawn run (14.50 days) were observed in ferrous sulphate 1.0% which was statistically at par with ferrous sulphate 1.2% (15.25 days). While maximum days for spawn run (20.25 days) were observed in control at par copper sulphate 1.0% (18.20) which was statistically. Minimum days for pin head formation (18.26 days) were observed in ferrous sulphate 1.0% which was statistically at par, with ferrous sulphate 1.2% (19.00 days). Maximum days for pin head formation (24.16 days) were observed in control which was statistically higher than all other treatment. Minimum days for first harvesting (22.30 days) were observed in ferrous sulphate 1.0% which was statistically lower than all other treatment. While maximum days for first harvesting (28.25 days) was observed in control which was statistically higher than all other treatment. The Maximum numbers of fruiting bodies (135.00) were observed in ferrous sulphate 1.0%. This was significantly higher than all other treatment. While minimum number of fruiting bodies (94.00) were observed in control. This was significantly lower than all other treatment. Maximum average weight of fruiting bodies (19.25 gm) was observed in zinc sulphate @ 1.2%. This was significantly higher than all treatment. While minimum average weight of fruiting bodies (14.00 gm) was observed in ferrous sulphate 1.0% which was significantly lower than all other treatments.

In case of *P. sajor-caju* maximum yield was observed in ferrous sulphate 1.0% (790.33 gm/kg of dry substrates with 79.03% biological efficiency.) which was followed by ferrous sulphate 1.2% (790.00 gm/kg of dry substrates with 79.00% biological efficiency). While minimum yield was observed in control (512.33 gm/kg dry substrates with 51.23% biological efficiency.) which was followed by zinc sulphate 1.2% (679.33 gm/kg of dry substrates with 67.93% biological efficiency) which was statistically lower than all other treatment minimum days for spawn run (18.15 days) were observed in ferrous sulphate 1.0% which was statistically at par with ferrous sulphate 1.2%, (18.60 days). While maximum days for spawn run (22.20 days) were observed in control which was statistically at par with zinc sulphate (20.22 days) which was statistically the minimum days for pin head formation (21.26 days) were observed in ferrous sulphate 1.0% which was statistically at par, with ferrous sulphate 1.0% and zinc sulphate 1.2% (22.00 days). Maximum days for pin head formation (25.16 days) were

observed in control which was statistically higher than all other treatment. The minimum days for first harvesting (24.20 days) were observed in ferrous sulphate 1.0% which was statistically lower than all other treatment. While maximum days for first harvesting (28.25 days) were observed in control was statistically higher than all other treatment. The Maximum numbers of fruiting bodies (137.33) were observed in ferrous sulphate 1.0%. This was significantly higher than all other treatment. While minimum number of fruiting bodies (114.00) was observed in control. This was significantly lower than all other treatment. The maximum average weight of fruiting bodies (25.00 g) was observed in ferrous sulphate 1.0%. This was significantly higher than all treatment. While minimum average weight of fruiting bodies (20.26 gm) was observed zinc sulphate 1.2% .Which was significantly lower than all treatment.

The results were in accordance with the findings of Bhadana, N. K. *et al.*, (2022) [1] studied the effect of micro-

nutrient on yield of *Pleurotus djamor* Maximum DFSR, DFFH and average weight was obtained in ferrous sulphate. The results were in accordance with the findings of Kachroo *et al.*, (1997) [5] studied the effect of micro-nutrient on yield of *P. sajor-caju*. Maximum yield was obtained in magnesium sulphate (187.22 gm), followed by zinc sulphate (176.22 gm) and ferrous sulphate (173.22 gm). Low yield was obtained in copper sulphate.

Huang *et al.* (2003) [3] studied the effect of various source of carbon (brown sugar, fructose, lactose, glucose, sucrose, starch and maltose), nitrogen [wheat bran, yeast cream, beef cream, peptone, (NH₄)₂SO₄, NH₄Cl and (NH₄)₂CO₃] and inorganic salts (K₂SO₄, MgSO₄, CaSO₄, MnSO₄ and FeSO₄) on the mycelial growth of *P. ostreatus*. The mycelial growth was more pronounced with brown sugar as the carbon source, wheat bran as the nitrogen source and MgSO₄ as the inorganic salts source. The most suitable culture medium for *P. ostreatus* consisted of various nutrient sources, including 3 per cent wheat bran, 1 per cent brown sugar, 1 per cent MgSO₄ and 10 mg vitamin B1 per litre.

Table 1: Effect of different inorganic supplements on spawn growth of *Pleurotus* spp. (*P. djamor* and *P. sajor-caju*)

S. No	Treatments	Dose (%)	Spawn growth (mm)						15 th day Growth rate (mm/day)	
			5 th Day		10 th Day		15 th Day		<i>P. djamor</i>	<i>P. sajor-caju</i>
			<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>		
1	Ferrous sulphate	0.5	35.00	37.66	50.66	57.33	86.00	90.00	5.73	6.00
2	Ferrous sulphate	1.0	34.00	37.33	49.33	56.00	82.66	87.66	5.51	5.84
3	Zinc sulphate	0.5	36.33	37.66	56.00	56.66	90.00	90.00	6.00	6.00
4	Zinc sulphate	1.0	36.00	35.00	56.00	55.00	87.66	82.66	5.84	5.51
5	Magnesium sulphate	0.5	35.66	36.66	56.33	55.00	90.00	88.33	6.00	5.88
6	Magnesium sulphate	1.0	35.00	35.00	51.66	52.00	86.33	84.33	5.75	5.62
7	Control		28.00	34.66	48.33	49.66	75.00	67.66	5.00	4.51
	CD at 5%		4.50	1.59	4.53	2.50	3.34	4.89	-	-
	SE (M)		1.46	0.51	1.48	0.81	1.09	1.59	-	-

Average of three replications

Table 2: Effect of different inorganic additives on yield of *Pleurotus* spp. (*P. djamor* and *P. sajor-caju*)

S. No	Inorganic supplement (%)	DFSR		DFFP		DFFH		NOFB		Yield (g/kg dry substrate)		Average weight (gm./FB)		Biological efficiency (%)	
		<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>	<i>P. djamor</i>	<i>P. sajor-caju</i>
		1.	Ferrous Sulphate @ 1.0%	14.50	18.15	18.26	21.26	22.30	24.20	135.00	137.33	462.66	790.33	14.00	25.00
2.	Ferrous Sulphate @ 1.2%	15.25	18.60	19.00	22.00	23.00	26.18	126.00	133.00	456.66	790.00	18.00	22.26	45.66	79.00
3.	Copper Sulphate @ 1.0%	18.20	19.15	21.00	22.25	23.10	25.20	120.00	125.00	438.33	788.66	17.00	24.16	43.83	78.86
4.	Copper Sulphate @ 1.2%	17.10	19.52	21.18	23.65	24.95	27.10	112.00	123.33	424.00	783.33	18.66	24.85	42.40	78.33
5.	Zinc Sulphate @ 1.0%	17.52	20.22	21.00	23.96	24.20	25.20	110.00	120.33	422.00	710.00	17.55	22.50	42.20	71.00
6.	Zinc Sulphate @ 1.2%	17.25	19.00	22.28	22.00	26.00	27.00	98.00	116.66	418.33	679.33	19.25	20.26	41.83	67.93
7.	Control	20.25	22.20	24.16	25.16	28.25	28.25	94.00	114.66	409.00	512.33	19.00	22.00	40.90	51.23
	CD at 5%	2.91	2.38	2.41	1.92	2.91	2.46	3.27	5.29	9.49	15.19	-	-	-	-
	SE (m)	0.95	0.78	.078	0.63	0.95	0.88	1.06	1.72	3.09	4.96	-	-	-	-

Average of three replications

DFSR= Days for spawn run, DFFP = Days for pinhead formation, DFFH= Days for first harvesting, NOFB=Number of fruiting bodies

Conclusion

Mushroom farming in India is gaining popularity due to its low input and potential to significantly change the rural economy. Oyster mushroom cultivation is becoming more profitable and popular in developing countries due to its easy and cheap cultivation technology, high nutritive value, good shelf life, distinctive taste, abundant availability of agro-wastes, and suitability to varying agro-climatic conditions. A study conducted in 2018 at the Mushroom Laboratory Department Plant Pathology, S. V. P. University of Agriculture and Technology, Meerut, India, focused on the growth and yield of *Pleurotus* spp. against various

inorganic additives in a laboratory and crop room. The study involved using wheat straw as a substrate for spawning *Pleurotus* spp. sporophores, maintaining a temperature of 22 to 26 °C and relative humidity of 80 to 90 percent. The study investigated the effects of inorganic supplements on the growth of *P. djamor* and *P. sajor-caju* oyster mushrooms. Results showed that Zinc sulphate 0.5% and Magnesium sulphate 0.5% had the highest spawn growth on the 5th, 10th, and 15th days. Ferrous sulphate 1.0% had the highest yield (462.66 gm/kg of dry substrates with 46.26% biological efficiency), followed by zinc sulphate 1.2% (456.66 gm/kg of dry substrates with 45.66% biological

efficiency). The study also found that ferrous sulphate 1.0% and zinc sulphate 1.2% were the most effective in increasing the yield of *P. sajor-caju*, with varying biological efficiency. Oyster mushroom cultivation is becoming more profitable in developing countries due to its easy and cheap cultivation technology, high nutritive value, good shelf life, distinctive taste, abundant availability of agro-wastes, and suitability to varying agro-climatic conditions. In our studies found that zinc sulphate 0.5% and magnesium sulphate 0.5% had the highest spawn growth.

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