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Research Article

STUDIED ON THE IMPROVEMENT OF SPAWN PRODUCTION BY SUPPLEMENTATION OF DIFFERENT SUGARS AND ITS SPAWN EFFECTS ON YIELD OF OYSTER MUSHROOMS *(Pleurotus djamor)*

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Abstract- Spawn is an essential component of mushroom production and it has a big effect on the sporophores production. During the mushroom session, production of quality spawn in minimum days and availability for planting beds of mushroom is a big challenge for mushroom growers. The present study was conducted with the aim to find out the most favorable sugar for the improvement of spawn quality and production in minimum days and its effect on yield and growth of Oyster mushroom (*Pleurotus djamor*). In the present Studyfive different sugars viz. Dextrose, Maltose, Starch, Sucrose and Glucose were mixed as a supplement with wheat grain for spawn production and effects of its spawn on the yield of sporophores and growth of *Pleurotus djamor* were also observed. The results obtained during the present investigation, in the spawn production maximum mycelial growth (98.00 mm) was found in glucose on 20th days and the mycelium was thick then others while in case of sporophores production maximum yield (613.33g/Kg of dry substrate with 61.33% B.E.) and minimum days for first harvesting (23.00 days) were observed in glucose spawn. Based on the results obtained, for production of *Pleurotus djamor* spawn and sporophores yield, glucose would be recommended most effective sugar for spawn and sporophores production useas supplement in Wheat grain.

Keywords- *Pleurotus djamor,* Mushroom, Spawn, Sugars, Supplements, Yield, Glucose.

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Introduction

Mushroom cultivation is a profitable agribusiness world-wide. There are more than 5000 mushroom varieties could be employed for foods and medicines. In the fungal classification system proposed by Ainsworth and followed by J. Webster [1], almost edible mushrooms are members of the subdivision Basidiomycotina and Ascomycotina [2]. Mushrooms are a good source of protein, vitamins and minerals and are known to have a broad range of uses both as food and medicine. A high nutritional value of oyster mushrooms has been reported with protein (25-50%), fat (2-5%), sugars (17-47%), mycocellulose (7-38%) and minerals (potassium, phosphorus, calcium, sodium) of about 8-12% [3].

The cultivation of edible mushrooms has become an attractive economic alternative over past few years, mainly due to increase in its demand and market value [4, 5]. Oyster mushrooms are one among the cultivable varieties. They are wide spread in temperate zones, can grow at moderate temperature and are suitable to grow in most places in India [5,6].Oyster mushrooms are grown from hyphae (threadlike filaments) that become interwoven into mycelium and propagated on a base of steam sterilized cereal grain usually Wheat grains. This mycelium-impregnated cereal grain is called spawn and is used to inoculate mushroom substrate [7]. Failure to achieve a satisfactory harvest may often be traced to unsatisfactory spawn used [8].

In other words spawn comprises mycelium of the mushroom and a supporting medium which provides nutrition to the fungus during its growth. The propagating material used by the mushroom growers for planting beds is called spawn. The spawn is equivalent to vegetative seed of higher plants [9]. In mushroom growing technology, the inoculums are known as the 'spawn'. Spawn is a medium that is impregnated with mycelium made from a pure culture of the chosen mushroom strain. Spawn production is a fermentation process in which the mushroom mycelium will be increased by growing through a solid organic matrix under controlled environmental conditions [10, 11].

The yield and quality of mushroom produced is determined by three factors: the genetic makeup of the mushroom strain, the environmental conditions in which the mushroom is grown and the physiological and nutritional requirements of different strains. Attempts on identification of potential strain and cultivation technology has been made till today but further research still needs to be carried out in the area such as, improvement in new hybrid strain, use of organic supplementation in cultivation, evaluation of locally available cheap grains for spawn, morphological characterization etc. Keeping in view the above points of importance and possibilities of cultivating Oyster mushroom in the rural as well as urban areas of the country, the present investigation was carried out with objectives to evaluate the beneficial sugars supplement for production of spawn as well enhance the yield of *Pleurotus djamor*.

Material and Methods

Experimental site

The experiments were conducted during 2014-2015 in Mushroom Laboratory Department Plant of 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 djamor* 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^{0}C for about a week, again sub cultured on PDA and then stored in a refrigerator at 5-10^{0}C for further use [12].

Spawn Production and adding of sugars

In present study, five different type sugars viz. Dextrose, Maltose, Starch, Sucrose and Glucose @ 1.2% were mixed as a supplement 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^{0}C. The cooled grains were then mixed with 1.2 percent commercial grade gypsum ($CaSO_4$) and 0.3 percent calcium carbonate ($CaCO_3$). Gypsum prevents the sticking of wheat grains together and calcium carbonate maintains the pH 5.5 - 7.5. The grains were filled up to (100 mm) in the bottle in three replicates. The bottles were plugged with non-absorbent cotton and covered with butter paper. These bottles were then sterilized at 121^{0}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. Before the inoculation pre balanced by electric balance and sterilized by autoclave (10 lb pressure for 15 minutes)[13]sugars were mixed in bottle under aseptic condition in laminar flow chamber. The spawn bottles were incubated without shaking at 24±1^{0}C in B.O.D incubator and observations were recorded on 5th, 10th, 15th and 20th day till to completely cover by mycelial growth in bottles.

Substrate Preparation:

Wheat straw was used as substrate for this experiment. It was soaked (10kg wheat straw/100liter water) in a tank with solution of Carbendazim (8gm/100liter water) + Formalin (120ml/100liter water) for 18 hr (tank should be covered with polythene sheet to prevent the evaporation of formalin) [14] 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 djamor* (preparation method described under spawn production)wasmixed in Wheat straw (substrate) @ 4 percent per kg on dry weight basis and 3kg 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. The observations were recorded as total yield (gm. /kg dry straw) and minimum days for spawn run (DFSR), minimum days for first harvesting (DFFH), number of lobe per beg (NOL), number of fruiting body per beg (NOFB) and maximum average weight of fruiting body (g/FB).

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 CO_2 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.

Harvesting

The sporophores of *P.djamor* were harvested after the maturity. Before the harvesting sporophores were irrigated for keep it fresh. The yield obtained in 7 weeks harvesting period were compared with each other. After first harvesting begs were scraped and remain without irrigation for three days and then again irrigated after pinhead initiation. Same process was follow after second harvesting. Adequate ventilation in the crop room was provided by opening the doors and windows at night for a short duration during cropping.

Statistical analysis

The Complete randomized design (CRD) was applied and the data thus obtained were analyzed statistically. Analysis of variance (ANOVA) technique and critical difference (CD) was calculated at five percent level of significance for comparison with other treatment [15].

Result and Discussion

In the present investigation of different sugars supplements for spawn and its spawn effects on sporophores production results shows that:

Effect of different sugars on spawn's production

In the present study five different sugars mixed in wheat grain viz. glucose, dextrose, maltose, sucrose and starch. The results revealed that among all the five sugar, maximum mycelial growth (98.00 mm) was found in glucose on 20th days which was significantly higher to all other treatments and it was followed by mycelial growth in sucrose (86.67 mm) and maltose (78.33mm) While dextrose (76.67) was significantly at par with maltose. The least mycelial growth of the *P. djamor was* recorded in starch (70.00 mm), followed by wheat grain without sugar i.e. control (59.33 mm) respectively on 20th days which was significantly lower than all other treatments. Results are shown in [Table-1], [Plate-1] and [Fig-1].

These results were found in proximity with the research findings of of Fallal [16] evaluated the effect of carbon source from sugars (glucose, fructose, maltose, lactose, glactose, raffinose, and inositol) on the growth of *P. columbines* and *P. pulmonarius*. The best carbon was obtained from sucrose and glucose for *P.*

pulmonarious and *P. columbines*. The least growth of *P. pulmonarious* and *P. columbinus* was found in the presence of lactose and glactose, respectively. Saurabh [17] determined that, the species of *P. flabellatus* were grown in five sugar mix in wheat grain *viz*. Fructose, Maltose, Sucrose, Starch and Glucose. The results revealed that among all the five sugar, maximum mycelial growth (90.00 mm) was found in glucose followed by maltose (85.50 mm). Bhadana [18] determined that, effect of different sugars on two species of *Pleurotus* spp. (i.e. *P. florida and P. djamor,*). The results revealed that, maximum mycelial growth of *P. florida* and *P. djamor* was found in glucose.

Plate-1 Effect of different sugars on spawn growth of oyster mushroom (*P. djamor).*

1. Glucose, 2. Sucrose, 3. Maltose, 4. Dextrose, 5. Starch, 6. Control.

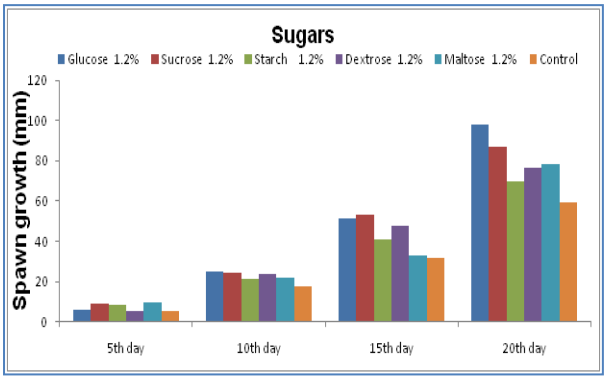

Fig-1 Effect of different sugars on spawn growth (mm) of oyster mushroom (*P. djamor)*

Effect of different sugars spawns on sporophores yield of *P. djamor*

In case of effects of different sugars added spawn,the data revealed that, maximum yield (613.33g/Kg of dry substrate with 61.33% B.E.) was observed in glucose spawn which was significantly similar with starch spawn. It was followed by sucrose and maltose spawn (500.00 g/kg,500.00g/kg of dry substrate with 50.00, 50.00% B.E.). The minimum yield was recorded in control spawn (without sugars) (400.00g/Kg of dry substrate with 40.00% B.E.) which was significantly lower than all other treatments.

The minimum days for spawn run (18 days) observed starch, which was statistically at par with all other spawns except to dextrose and control spawn (without sugars). The maximum days for spawn run (25.00 days) were observed in control spawn (without sugars) which was significantly higher than all other treatments. The minimum days for first harvesting (23.00 days) was observed glucose, which was statistically similar with starch and maltose. The maximum days for first harvesting (30.00 days) was observed in control spawn (without sugars) which was significantly higher than all other treatments.

The highest number of fruiting body (29.20) was observed in dextrose spawn, which was significantly higher than all other treatments. The minimum number of fruiting body (17.00) was observed control spawn (without sugars) which was significantly lower than all other treatments. The maximum average weight of fruiting body (25.98) was observed in glucose spawn which was significantly at par with starch. The minimum average weight of fruiting body (16.78) was observed in dextrose was significantly lower than all other treatments.

The highest number of lob (81.60) was observed in dextrose spawn which was significantly higher than all other treatments. The minimum number of lob (56.40) was observed was observed control spawn (without sugars) which was significantly lower than all other treatments. The highest pileus length as well as width (9.80 cm and 9.50 cm) was observed in sucrose which was significantly higher than all other spawn. The minimum pileus length and width was observed in control spawn (without sugars) (6.33 cm and 9.00 cm) which were significantly at par with maltose, dextrose and glucose. The maximum days for cropping period (74.67 days) were observed in control spawn (without sugars) which was statistically similar with maltose and glucose. The minimum days for cropping period (69.00 days) were observed in sucrose which was statistically similar with starch and dextrose (69.00). Results are shown in [Table-2] and [Fig-2].

These results were found in proximity with the research findings of the results were in accordance with the findings of Gbolagade [19] studied that, the effect of different carbon sources such as; (glucose, fructose, maltose mannose sorbose) on the yield of oyster mushroom (*Pleurotus flabelatus*). Result revealed that the best yield production followed in order by fructose, mannose, and sorbose. Mohamed [20] studies that, the effect of various carbon sources such as glucose, fructose, maltose, lactose, sucrose were tested on yield of oyster mushroom (*Pleurotus ostreatus)*. Result revealed that, the highest yield on glucose, followed by fructose.

Table-1 *Effect of different sugars on spawn growth (mm) of oyster mushroom (P. djamor)*

Sugars	Dose (%)	5th day		10th day		15thday		20th day	
		spawn growth (mm)	growth rate (mm/day)	spawn growth (mm)	growth rate (mm/day)	spawn growth (mm)	growth rate (mm/day)	spawn growth (mm)	growth rate (mm/day)
Glucose	1.2	6.00	1.20	25.33	2.53	51.67	3.44	98.00	4.90
Sucrose	1.2	9.00	1.80	24.33	2.43	53.33	3.55	86.67	4.33
Starch	1.2	8.67	1.73	21.67	2.16	41.00	2.73	70.00	3.50
Dextrose	1.2	5.67	1.13	23.67	2.36	47.67	3.17	76.67	3.83
Maltose	1.2	9.67	1.93	22.00	2.20	33.00	2.20	78.33	3.91
Control (without sugar)	_	5.33	1.06	18.00	1.80	31.70	2.11	59.33	2.96
SE	_	1.17	_	1.01	_	1.12	_	1.57	_
CD at 5%	_	2.57	_	2.24	_	2.47	_	3.47	_

Table-2 *Effect of different type sugars additive spawn on spawn run, cropping period and yield of oyster mushroom (P. djamor)*

Sugars	DFSR	DFFH	DFCP	NOFB	NOL	Pileus length (in cm)	Pileus width (in cm)	Yield (g/kgdry Substrate)	Average Weight /FB	Biological efficiency (%)
Glucose	18.00	23.00	72.00	23.60	62.00	7.33	6.00	613.33	25.98	61.33
Sucrose	19.00	26.00	69.00	22.26	68.80	9.80	9.50	500.00	22.46	50.00
Starch	18.00	23.00	69.00	23.60	62.40	8.00	8.00	610.00	25.84	61.00
Dextrose	21.00	27.00	69.00	29.20	81.60	7.67	6.50	490.00	16.78	49.00
Maltose	18.00	23.00	74.00	25.20	59.13	7.00	7.00	500.00	19.84	50.00
Control (without sugars)	25.00	30.00	74.67	17.00	56.40	6.33	9.00	400.00	23.52	40.00
SE	1.33	1.29	1.34	0.96	0.77	0.74	0.66	7.69	0.90	_
CD at 5%	2.93	2.84	2.96	2.12	1.69	1.64	1.46	16.95	1.98	_

DFSR= Days for spawn run, DFCP= Days for cropping period, DFFH= Days for first harvesting, NOFB= Number of fruiting body, NOL- Number of lob

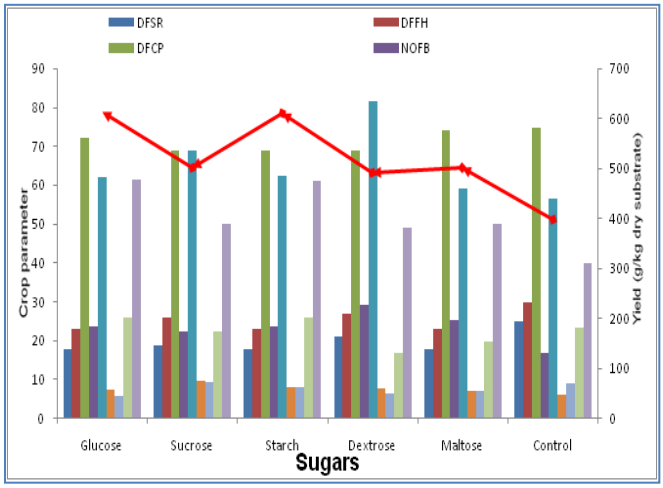

Fig-2 Effect of different type sugars additive spawn on spawn run, cropping period and yield of oyster mushroom (*P.djamor*).

Conclusion

In the mushroom cultivation spawn is an essential component, which affects to yield of mushroom hence a study, was conducted to determine the effect of different sugars supplementation on mycelial growth in spawn and enhance the yield and growth of Oyster mushroom. The maximum mycelial growth (98.00 mm length) was found in wheat grain added with 1.2% glucose on 20th days while in case of yield; maximum yield was recorded from the spawn added with glucose and thus it's recommended for spawn production and for cultivation of *Pleurotus djamor* as sugar supplement (in Wheat grain) to be use.

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Conflict of Interest: None declared

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