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The effect of nutritional supplement addition and use of casing overlay to substrate on yield of oyster mushroom (*Pleurotusflorida*)

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Abstract

Improving yield, biological efficiency (BE) and number of pinhead formation in *Pleurotusflorida* where achieved by nutritional supplement addition and use of casing overlay to substrate. In the present study utilization of soybean meal at two levels (2.5%- 5 % dry substrate weight) and Hogland solution (Hogland, ½ Hogland) without the use or with avail of casing overlay to substrate and with control substrate were evaluated. The results indicated that the addition of nutritional supplement and use of casing overlay to substrate had significant effect on yield and pinhead formation of oyster mushroom. Among the treatments, substrate received Hoagland solution (H, ½ H) without use of casing overlay had the lowest number of days to pinhead formation (20.5 days). The maximum first yield (492.96 g/g) was observed in the use casing supplemented with 2.5% of soybean meal. Casing combined with ½ Hogland solution had the highest total yield (1121. 71 g/g), and biological efficiency (140.215). In general, It seems that application of Hogland solution for mushroom cultivation was very helpful to obtain high yield and high quality.

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Introduction

Oyster mushrooms belong to the genus *Pleurotusin* the family of *Pleurotaceaeare* one of the most well-known edible mushrooms. Kues and liu (2000) reported that the *Pleurotus*species are popular mushrooms in the world. Pleurotus mushrooms same as oyster mushrooms are primary decomposers of hardwood trees and are found worldwide. *P. florida* is widespread in temperate, subtropical and tropical zones. It is similar in appearance and was considered as subspecies of *P. ostreatus*. Some modern mycologists are inclined to regard it as another species with different color and different temperature requirements.

Shukla and Biswas (2000), Mane *et al.,* 2007 stated that due to the higher biological efficiency, low cost of production and easy technology to produce oyster mushroom (*Pleurotusflorida*) cultivation is gaining popularity in different parts of the world. Oyster mushroom growers use wide range of substrate materials as oyster mushroom can utilize various agro- wastes thanks to its enzymes. That is to say, oyster mushroom is a white rot fungus that uses lignin and cellulose together as its carbon source. Therefore, any type of organic matters containing lignin and cellulose can be used for oyster mushroom production as substrates, and this includes almost all agricultural wastes. Possible substrate materials are sunflower seed hulls, rice/wheat straw, bean, sugarcane biogases, rubber tree sawdust, groundnut shells, cotton waste, cottonseed hulls, coco lumber sawdust, coffee pulp, corncobs, paper, water hyacinth, water lily, cocoa shell waste, coir and others.

Many cultivated edible mushrooms for example *Agaricusbisporus*, *Lentinulaedodes*, and *Pleurotu-sostreatus* have more than one break or flush and growers typically harvest three breaks (Royse, 2001; Velazquez-Cedeno *et al*., 2002; Royse *et al*., 2008 Alma *et al*., 2009). Important factor for growth the mushroom are substrate and supplementation, except substrate and supplementation include moisture content, temperature, pH and light intensity (Ibekwe *et al*., 2008; Stamet, 1993). Nutrient depletion occurs in button mushrooms substrates due consecutive harvest and Followed by the product decreases (Schisler, 1964; Royse *et al*., 2008) Therefore supplement addition to the substrates is a strategy to improve yield and biological efficiency at early or later stages of the production cycle (Schisler and Sinden, 1962; Royse *et al*., 2004; Rodriguez Estrada and Royse, 2007), that one important method in commercial cultivation of *A. bisporus* addition supplement at spawning or casing with slow-release nutrients(Alma *et al*., 2009). Use a casing overlay makes more than one break and at least reduce the loss of substrate moisture content that for where environmental controls are difficult or lacking or where production is outdoors is a good way (Tan *et al*., 2005; Oei, 2006; Rodriguez Estrada and Royse, 2008).

In this work, to increase biological efficiency and yield of *P. florida* using casing overlay and addition of popular mineral nutrient solution (Hogland) supplement to the substrate also evaluated the effects of these two factors on the number of days to pinhead formation.

Materials and methods

Experimental design

The experiment included 10 treatments, treatments 1-4 include nutritional supplement addition without the use of casing overlay, treatments 5-8 include nutritional supplement addition whit use casing overlay, treatments 9 and 10 served as two controls (Table1). Each treatment was replicated in four plastic bins. The plastic bins were assigned at random in three tiers from top to bottom in the cropping room level of confidence.

Spawn

Spawn of *P. florida* was purchased from the company BIBI GOLSHAR KARJ.

Substrate preparation

Mushrooms were produced on a substrate that contained :wheat straw, the material is milled to a length of about 3 - 4 cm. moistened substrate

autoclaved at 120˚C for 120 min. 800 g of cooled substrate was inculcated with 20 g (± 0.1 g) of spawn. Plastic bins(30 cm long, 20 cm wide, 8.5 cm deep) sterilized with alcohol 70 %. The running was in growing room under controlled conditions. The room was darkness and temperature kept 24˚C in this phase. After 25 days of running the room temperature was lowered to 16˚C with cycle of 8h light/ 16h dark. The relative humidity was kept at 85 - 90 %.

Table 1. The treatments used in this study.

Treatment number	Treatment designation	Casing layer	Supplement applied time	
1	NC/S/AS (S, 2.5%)	No	Spawning	Soybean meal (2/5%)
2	NC/S/AS (S, 5%)	No	Spawning	Soybean meal (5%)
3	NC/S/AS (H)	No	Spawning	Hogland
4	NC/S/AS/ (½ H)	No	Spawning	½ Hogland
5	C/S/AC (S, 2.5%)	Yes	casing layer	Soybean meal (2/5%)
6	C/S/AC (S, 5%)	Yes	casing layer	Soybean meal (5%)
7	C/S/AC (H)	Yes	casing layer	Hogland
8	C/S/AC (½ H)	Yes	casing layer	½ Hogland
9	NC	No	-	-
10	C	Yes	-	-

NC: non-cased; NC/S/AS (S ,2.5%): non-cased, supplemented (Soybean meal added 2.5% dry substrate weight) at spawning; NC/S/AS (S, 5%): non-cased, supplemented (Soybean meal added 5% dry substrate weight) at spawning; NC/S/AS (H): non-cased, supplemented (Hogland) solution at spawning; NC/S/AS (½ H): non-cased, supplemented (½ Hogland) at spawning; C: cased; C/S/AC (S, 2.5%): cased, supplemented (Soybean meal added 2.5% dry substrate weight) at casing layer; C/S/AC (S, 5%): cased, supplemented (soybean meal added 5% dry substrate weight) at casing layer; C/S/AC (H): cased, supplemented with (Hogland) solution at casing layer: C/S/AC (½ H): cased, supplemented (½ Hogland) solution at casing layer.

Nutritional supplement addition

Nutritional supplementation used in this experiment included soybean meal at two levels (2.5% and5 % of dry substrate weight) and Hogland solution (Hogland, ½ Hogland) that autoclaved at 120˚C for 120 min. Treatments 1-4 (NC/S/AS (S, 2.5%), NC/S/AS (S, 5%), NC/S/AS (H), NC/S/AS/ (½ H) respectively, (Table 1) were supplemented at time of substrate preparation and kept without casing. Because the substrate was inoculated the day after it was prepared, it was referred as "supplementation at spawning" (AS). The substrate was sterilized, spawned, supplemented and transferred to the culture room. Bins for treatments 5-8 and 10 (C/S/AC (S, 2.5%), C/S/AC (S, 5%), C/S/AC (H), C/S/AC (½ H), C respectively,) (table 1) were cased with vermicompost casing that was overlaid (3 cm) on the surface of exposed. These treatments were supplemented at time of adding casing overlay 10 days after spanning, and it was referred as "supplementation at casing layer".

Evaluation methods

Number of days to pinhead formation, first and total yield, Biological efficiency (BE)

Number of days to pinhead formation, yield, BE were determined for all treatments. Total weight of all the mushrooms harvested in the first picking were measured as first yield of mushroom and total yield was expressed as fresh mushroom weight (g) per bin; biological efficiency (BE) were estimated as the ratio of mushroom fresh weight to dry substrate weight and expressed as a percentage (Chang *et al.,* 1981).

Statistical analyses

Analysis of variance (ANOVA) and mean comparison were performed using IBM SPSS statistical software (version 20). All data (number of days to pinhead formation, yield and biological efficiency) were analyzed using the general linear models procedure of SPSS. Differences among means were tested for significance <0:01 by Duncan multiple range test. Charts were plotted using Excel software.

Results

Number of days to pinhead formation, first and total yield, BE

First mushrooms harvested began 28 days after spawning. Number of days to pinhead formation, First and total yield and BE were significantly affected by casing layer and supplementation (P< 0.01) (Table 2). The lowest days to pinhead formation (20.5 days) were observed for treatments NC/ S/ AS (H) and NC /S/ AS (**½** H) (non- cased, supplemented (Hogland) at spawning, non- cased, supplemented (**½** H) at spawning, respectively), while highest number of days to pinhead formation (35/25 days) were observed for C/S/AC (S, 5%) cased, supplemented (soybean meal added 5% dry substrate weight) at casing layer (Fig.1-a).

Table 2. Means and standard deviation for number of pinhead formation, first yield, total yield and BE for production of *Pleurotus florida* influenced by substrate supplementation and application of casing layer.

Treatment [a]	Number of pinhead formation	First yield [b]	Total yield [b]	BE [b]
Control	25 ± 0.816	122.96 ± 18.95	191.815 ± 11.95	23.977
NC/S/AS (S, 2.5%)	25.25 ± 0 .957	152.45 ± 41.25	934.675 ± 46.23	116.834
NC/S/AS (S, 5%)	28.75 ± 0.5	91.27± 19.70	266.342 ± 17.62	33.293
NC/S/AS (H)	20.25± 0.577	147.39 ± 37.41	959.58 ± 19.74	119.948
NC/S/AS (1/2H)	20.5± 0.577	152.41 ± 45.69	685.414 ± 14.1	85.677
C/S/AS (S, 2.5%)	31.5 ± 1	429.96 ± 66.17	673.252 ± 21.79	84.156
C/S/AS (S, 5%)	35.25 ± 3.5	277.40 ± 14.14	292.76 ± 9.28	36.595
C/S/AS (H)	28.75± 0.5	65.193 ± 14.07	287.78 ± 9.76	35.972
C/S/AS (1/2H)	27± 0	59.05 ± 25.101	1121.718 ± 55.47	140.215

[a]NC/S/AS (S ,2.5%): non-cased, supplemented (Soybean meal added 2.5% dry substrate weight) at spawning; NC/S/AS (S, 5%): non-cased, supplemented (Soybean meal added 5% dry substrate weight) at spawning; NC/S/AS (H): non-cased, supplemented (Hogland) at spawning; NC/S/AS (½ H): non-cased, supplemented (½ Hogland) at spawning; C/S/AC (S, 2.5%): cased, supplemented (Soybean meal added 2.5% dry substrate weight) at casing layer; C/S/AC (S, 5%): cased, supplemented (soybean meal added 5% dry substrate weight) at casing layer; C/S/AC (H): cased, supplemented (Hogland) at casing layer: C/S/AC (½ H): cased, supplemented (½ Hogland) at casing layer.

[b]Means ± standard deviation indicates significant differences according to Duncan (P<0.01).

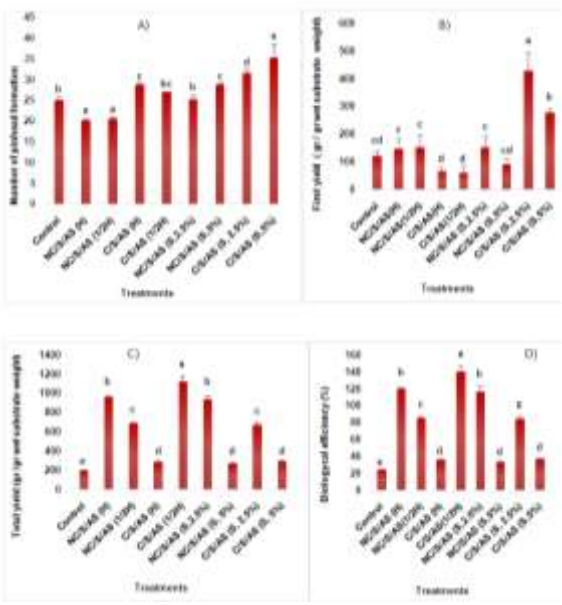

Fig. 1. Effects of treatments on the number of pinhead (A), first yield (B), total yield (C) and biological efficiency (D) of oyster mushroom.

First yield were higher (492.96 g) in cased, supplemented (Soybean meal added 2.5% dry substrate weight) at casing layer C/S/AC (S, 2.5%) compared to other treatments, while this parameter lowest (59.05 gr) were obtained for treatments cased, supplemented (Hogland and ½ Hogland) at casing layer(C/S/AC (H), C/S/AC (½ H)) (Fig 1-b). Highest total yield (112/71gr) and BE were obtained treatment C/S/AC (½ H) (cased and supplemented with ½ Hogland at casing layer) and lowest yield (191.81gr) (Fig. 1-c) and BE were observed in controls that yield and BE (Fig.1 d4).

Discussion

The results from the experiments mentioned above indicate that cased treatments May results delay in pinhead formation in comparison with non- cased substrate. In oyster mushroom the primordial

initiation mainly occurs on the 24th-30th day after spanning (Khanna *et al.*, 1992). Naraian *et al.* (2009) reported primordial initiation on the 20.2nd – 35.1th day in *Pleurotus florida* on corn cob substrate that supplemented with different nitrogen rich resources. In the present study, NC/S/AS (H) treatment has best results in shorten duration in term of the pinhead formation. Overall performance(1121.718 g) and BE (140.215) were highest on C/S/AS (1/2 H) treatment that harvested mushroom from this substrate were 5.8timehigher than the other treatments. Biological efficiencies in oyster mushroom higher compared to other edible mushroom (Anderson, 1942). BE of *P. florida* obtained from cased/ supplemented (1/2 H) substrate in these experiments were significantly higher than values reported by Pradeep Kumar *et al.*(2000)obtained a maximum BE of 98.0% in *Ageratum* twigs followed by 90% on paddy straw. Yilis *et al.* (2003) reported that *Pleurotusostreatus,* grown on substrate wheat straw with soybean meal, gave the highest yield and BE. Alma and *et al.* (2007) observed 45.21 -125.7% BE of *Pleurotusostreatus*. Alma and *et al.* (2009) obtained BE of 35.9% to 53.1 % in non-cased treatments and 114.8% to 132.8 % in cased treatments. Patil *et al.* (2010) recorded on soybean straw 851.66 g/ kg, dry straw (85.16 % BE). The biological efficiency of the substrate indirectly denotes the suitability of the substrates for cultivation of particular strains of mushrooms (Ragunathana and Swaminathan, 2003). Nutritional supplement addition and use of casing overlay to substrate in other treatments except C/S/AS (1/2H) have not significant effect on yield and BE. This results may be agreement reports that casing soil should be of low nutritional content (Masaphy *et al.,* 1989) to benefit the formation of sporophore. Since casing soil is not practice commonly used in the commercial cultivation of oyster mushroom we need more studies in this filed.

The Hogland solution with different modification widely used in all over the world by vegetable growers, but as far as we know it is not used in mushroom production. In present study, result showed that Hogland solution for mushroom cultivation and obtained interesting and significant results. But in order to recommend it to the manufacturers its need to further studies and some other aspects.

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