

**EFFECT OF DIFFERENT NITROGEN SOURCES ON PLEUROTUS
CITRINOPILEUTUS SPAWN SPREAD AND FRUIT BODY FORMATION**

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Abstract

Pleurotus citrinopileatus, the golden oyster mushroom was cultured in different substrates such as: sawdust, bagasse, rice straw. The results showed that the spawn grows fastest in sawdust (0.78cm/day). Besides, the cultivation of golden oyster mushroom in sawdust supplemented with organic protein as rice bran, corn bran or inorganic nitrogen as urea, diammonium phosphate showed that supplementing 4% of corn bran or 3% diammonium phosphate is most appropriate. Conditions for growing mushrooms are 22-30°C, humidity 70-90%.

Keywords: the golden oyster mushroom, *Pleurotus citrinopileatus*, organic and inorganic protein.

1. INTRODUCTION

Vietnamese climate is well-suited for the growth of many fungi species which oyster mushroom is one of the most widely cultivated due to the high yield and variety of species. Golden oyster mushroom, *Pleurotus citrinopileatus*, is not only high nutritional but also medicinal value among them. Moreover, it has a source of immune-regulating antioxidants, anti-tumor, and anti-diabetic activity (Chen, 2009; Frimpong-Manso, 2011). Thus, the study of the effect of different protein sources on *Pleurotus citrinopileatus* is urgently needed to improve the process of mushroom cultivation for high yield.

2. MATERIALS AND METHODS

2.1 Materials

Fruiting body of *Pleurotus citrinopileatus* was provided by Bao Han mushroom farm at Long Khanh town, Dongnai province.

2.2 Methods

Substrates as sawdust, straw, bagasse were treated with lime water to 60% of humidity and heaping. After incubation, they were mixed with different proportions, packing 500 grams, then sterilizing at 121°C for 1 hour. Next, bags were inoculated seedling and incubated at 25°C. As the white mycelium were full of the bags, we conducted to irrigate and collect the fruit bodies. The temperature in planting house was about 22-30°C, diffused light 200-300 lux and humidity 70-90%.

2.2.1. Effect of some substrates on the spread of mycelium

Treatments were mixed in as follow: 100% sawdust (control), 25% sawdust + 75% straw, 50% sawdust + 50% straw, 75% sawdust +25% straw, 25% sawdust + 75% bagasse, 50% sawdust + 50% bagasse, 75% sawdust + 25% bagasse. Substrate bags were inoculated and incubated for 5 days, then recording the rate of spawn in the treatments and observing the fungal hyphae on each substrate.

2.2.2. Effect of different nitrogen sources on the spawn spread and fruit body formation

Selecting the substrate for the fastest rate of spawn in Experiment 1, next mixing with different nitrogen sources such as: organic protein (rice bran, corn bran) at the rate of 2%, 4%, 6%, 8%, 10%; inorganic nitrogen (urea and diammoni phosphate) at the rate of 1‰, 2‰, 3‰, 4‰,5‰. Substrate bags were inoculated and incubated for 5 days, then observed the rate of spawn. When the fungal hyphae spread full of the bags, they were watered to collect mushroom. The speed of spawn, fresh mushroom weight, and harvesting time of colecting fruit bodies were recorded.

2.2.3. Statistical analysis

Data were expressed as Means \pm SD, and the statistical significance of the differences between groups was evaluated by analysis of variance (ANOVA), and compared using least significant difference (LSD) at $p < 0.05$. The Pearson correlation was calculated to examine the relationships with 95% confidence intervals.

3. RESULTS AND DISCUSSION

3.1 Effect of some substrates on the spread of mycelium

The most remarkable results to emerge from the data is that the golden oyster mushroom spawn growing well in all substrates expressed its white and strong mycelia. Time and the

spawn speed in treatments were significantly different from the control (Table 1). Moreover, the speed of spawning in the control sample was the fastest 0.78cm per day, 1.4 times more than sample added 25% sawdust + 75% straw (slowest rate of 0.55cm per day). Supplementing 25% bagasse or 25% straw with sawdust made the growth of mushroom similar. According to Frimpong-Manso et al. (2011), rubber sawdust containing lots of cellulose, less hemicellulose and lignin, small particle structure, as well as good air permeability and moisture retention helped mushroom spawn spread quickly. In the other hand, bagasse and straws is rich in nutrients but it has fibrous structure, so fungal enzymes is difficult to hydrolyze the polymeric compounds of the substrate, which leads to mycelium growth slower than that of sawdust.

Table 1. Effect of some substrates on the spread of mycelium

Ingredient	Time of spreading mycelium (days)	Spawn speed(cm/day)
Sawdust 100 %	20.3 ^a	0.78 ^a
25% bagasse + 75% sawdust	23.2 ^b	0.69 ^d
50% bagasse + 50% sawdust	24.9 ^c	0.64 ^c
75% bagasse + 25% sawdust	24.2 ^c	0.66 ^c
25% straw + 75% sawdust	23.1 ^b	0.70 ^d
50% straw + 50% sawdust	26.3 ^d	0.61 ^b
75% straw + 25% sawdust	29.3 ^e	0.55 ^e

Data are expressed as mean ± standard deviation (SD) (P<0.05).

3.2 Effect of different nitrogen sources on the spawn spread and fruit body formation

After selecting the most suitable substrate as 100% sawdust for the fungal spawn in Experiment 1, we carried out adding different nitrogen sources to the medium, packing, sterilizing and incubating. The results showed that 100% of the bags were full of fruit bodies (Table 2).

3.2.1 Effect of organic nitrogen sources on the spawn spread and fruit body of golden oyster mushroom

Table 2. Effect of organic nitrogen sources (rice bran, corn bran) on the spawn spread and fruit body of golden oyster mushroom

The rate of bran supplemented (%)	Time of spreading mycelium (days)	Spawn speed (cm/day)	Time of creating fruit body	Mushroom weight (g)
Control	20 ^e	0.8 ^a	32.4 ^d	92.40 ^a
Rice bran 2%	19.3 ^d	0.82 ^b	27.0 ^b	102.60 ^b
Rice bran 4%	14.6 ^a	1.10 ^e	23.7 ^a	130.13 ^k
Rice bran 6%	14.4 ^a	1.11 ^e	24.0 ^a	121.00 ^g
Rice bran 8%	18.3 ^c	0.87 ^c	27.1 ^b	116.34 ^{ef}
Rice bran 10%	18.7 ^c	0.86 ^c	26.7 ^b	116.59 ^f
Corn bran 2%	17.0 ^b	0.94 ^d	23.9 ^a	106.59 ^e
Corn bran 4%	14.4 ^a	1.11 ^e	24.1 ^a	137.05 ^l
Corn bran 6%	17.0 ^b	0.94 ^d	26.4 ^b	126.76 ^h
Corn bran 8%	18.7 ^c	0.86 ^c	29.9 ^c	114.06 ^d
Corn bran 10%	18.3 ^c	0.87 ^c	30.7 ^c	115.13 ^{de}

Data are expressed as mean ± standard deviation (SD) (P < 0.05).

It is interesting fundamental to note that the spawn speed in culture supplemented 2% rice bran was the lowest (0.8 cm/day) and there was no difference compared to the control. The rate of rice bran supplemented from 4% to 6% in the medium helped the spawn speed reach the highest (1.11 cm/day), while increasing this ratio to 8% and 10%, the spawn speed decreased. Besides, the spread of mycelium in the medium supplemented with 4% corn bran gave the same result (1.1cm/day), and the medium supplemented with 2% or 6% gave lower results (0.94cm/day). According to Gibriel (1996), the growth of mycelium in medium containing glucose and sucrose was better than in other different carbon sources. However, too much glucose and sucrose led to the rapid development of unfavourable microflora competing food with fungus. Thus, adding rice bran 4% to 6% or corn bran 4% was suitable for yellow abalone mushroom *Pleurotus citrinopileutus*.

About creating fruit body time, all substrates supplemented bran gave results better than control samples, in which sawdust was supplemented with rice bran 4% to 6% or corn bran 2% to 4% for the earliest fruit body yield (8 days faster than control treatment). Meanwhile, increasing the rate of corn bran to 6%, 8%, and 10% or supplementing with rice bran 2%, 8%, or 10% in sawdust treatments also prolonged the fruit output. Besides, the development of fast and strong mycelium with dense structure accelerates fruit formation (Musieba, 2012). This explained that sawdust supplemented with suitable bran (4% to 6% rice bran or 4% corn bran) improved spawn speed and shortened fruit collection time. Moreover, the highest fruit

body weight (137.05g) was 1.5 times as much as the control in treatment adding corn bran at the rate of 4% while supplementing 6% corn bran or 4% rice bran gave lower fresh fruit body weight (126.76g and 130.3 g, respectively), 1.3 to 1.4 times higher than the control. The most remarkable result to emerge from the data is that mushroom in the treatment adding 4% corn bran was more bright yellow, thick and firm. Therefore, it is recommended to add 4% corn bran to collect better quality mushroom.

3.2.2 Effect of inorganic nitrogen sources (urea and diammonium phosphate (DAP)) on the spawn spread and fruit body of golden oyster mushroom

Table 3. Effect of inorganic nitrogen sources on the spawn spread and fruit body formation

The rate of bran supplemented (%)	Time of spreading mycelium (days)	Spawn speed (cm/day)	Time of creating fruit body	Mushroom weight (g)
Control	20.0 ^f	0.80 ^a	32.0 ^h	92.40 ^a
DAP 1‰	17.6 ^e	0.91 ^b	26.0 ^f	96.13 ^b
DAP 2‰	15.4 ^{ab}	1.03 ^{ef}	24.0 ^c	104.08 ^c
DAP 3‰	15.3 ^{ab}	1.05 ^{ef}	21.8 ^a	130.32 ^e
DAP 4‰	15.7 ^b	1.02 ^e	23.1 ^b	122.49 ^f
DAP 5‰	17.6 ^e	0.91 ^b	25.1 ^e	100.51 ^g
Urea 1‰	15.2 ^f	1.05 ^f	25.3 ^{ef}	117.25 ^h
Urea 2‰	17.0 ^c	0.94 ^c	27.3 ^g	119.78 ^d
Urea 3‰	0	0	-	-
Urea 4‰	0	0	-	-
Urea 5‰	0	0	-	-

Data are expressed as mean ± standard deviation (SD) (P<0.05).

As shown in Table 3, all substrates with DAP supplementation shortened the creating fruit body time, in which the medium supplemented with DAP 3‰ gave the shortest collecting mushroom time (21.8 days), 10 days earlier than control sample. Increasing or decreasing this ratio prolonged the fruiting time. On the other hand, urea supplementation at 1‰ and 2‰ also shortened the fruiting time (5-7 days earlier than the control samples) but it was still longer than DAP 1‰ supplement. For control samples, fresh mushroom weight was lower than all treatments adding inorganic nitrogen sources such as urea (1 to 2‰) and DAP (1 to 5‰). Addition of DAP at a rate of 3‰ in sawdust gave the highest fresh weight (130.32g), 1.4 times higher than that of the control treatment. However, when this ratio changed (increase or decrease), the fresh weight decreased.

3.3 Comparison of golden oyster mushroom spawn spread and fruit formation in medium supplemented different nitrogen sources

The results from Table 4 highlighted that there was significant difference in fruiting time and fresh mushroom weight except spawn speed when different nitrogen sources was added to the sawdust medium. In terms of creating fruit body time, supplementing DAP 3‰ shortened fastest time by only 21 days, on the contrary, at the rate of urea 1‰ gave the slowest time (25.3 days). The fresh mushroom weight in the medium supplemented with 4% corn bran was highest (137.05g), while it was lower than in sawdust added rice bran 4% (130.13g) and DAP 3‰ (130.32 g) and lowest in the medium added urea. Thus, in terms of fresh mushroom weight, the sawdust supplemented with 4% corn bran was appropriate for *Pleurotus citrinopileutus* mushroom.

Table 4. Ability to spread mycelium and take form fruit body of *Pleurotus citrinopileutus* in sawdust supplemented with different nitrogen sources

Additional components	Spawn speed (cm/day)	Time to create fruit body (day)	Fresh mushroom Weight (g)
Rice bran 4%	1.10	23.7	130.13
Corn bran 4%	1.10	24.1	137.05
DAP 3 ‰	1.05	21.08	130.32
Urea 1 ‰	1.05	25.3	117.25

4. CONCLUSION

100% sawdust with supplementing 4% corn bran or DAP 3‰ is best suited for the growth of golden oyster mushroom *Pleurotus citrinopileutus*.

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