BIBLIOGRAPHY

ANGIE C. JUAN. APRIL 2006. The Effects of Potato Dextrose Agar Composition and Light Supply on the Growth of Shiitake *(Lentinula edodes)* Pure Culture. Benguet State University, La Trinidad, Benguet.

Adviser: Janet S. Luis, PhD.

ABSTRACT

The potato dextrose agar (PDA) composition and light supply were evaluated to determine their effects on the growth and quality of shiitake pure culture. The study was conducted from November 2005 to February 2006 at the Plant Pathology Service Laboratory, Department of Plant Pathology, College of Agriculture, Benguet State University, La Trinidad, Benguet.

Based from the results of the experiment, the 3 days after inoculation (DAI) growth of the mycelia of shiitake on PDA gave the fastest mean mycelial growth (3.24 mm) and gave better growth when treated with continuous light at 3.00 mm which did not differed significantly from alternating light with darkness at 2.67 mm. The PDA concentrate costs P200 to produce a liter which is more expensive than PDA formulated from potato, dextrose and agar at P150.

At 14 DAI, the highest mean mycelial growth was observed from the PDA concentrate medium with 43.42 mm. Lighting did not affect mycelial growth.

In terms of the average mean daily mycelial growth, PDA concentrate gave the fastest mycelial growth with mean of 3.02 mm. Lighting had no effect on mycelial growth.

In terms of culture quality, concentrate PDA and PDA formulated from potato, dextrose and agar had abundant, filamentous, white and thick mycelia. The PDA formulated from potato, white sugar and gulaman bar had scanty, filamentous, creamy and thin mycelia.

TABLE OF CONTENTS

INTRODUCTION

Shiitake (*Lentinula edodes*) is a popular mushroom in the world. The shiitake, meaning "mushroom of the shii or oak tree" in Japan, is highly prized in the orient for its flavor and reported medicinal value. It is a major agricultural commodity in Japan, where about half the world's supply of shiitake is produced (Anonymous, 1997).

The shiitake is a traditional delicacy in Japan, Korea and China. For at least a thousand years, shiitake have been grown on logs outdoors in temperate mountainous region of Asia. To this day, shiitake is the most popular of the entire gourmet mushroom. Only in the past several decades have techniques involved for its rapid cycle cultivation indoors on supplemented, heat-treated sawdust-based substrates (Stamets, 1993).

Most shiitake are produced on natural logs but many farmers in the United State of America, Taiwan, China, Canada and Singapore are now producing shiitake on synthetic substrate composed mainly of oak sawdust in fully environment-controlled buildings providing conditions most favorable to the mushroom at their various stages of growth (Quimio, 1980).

This study was conducted to gather and provide some valuable information concerning the effects of potato dextrose agar (PDA) composition as culture medium and light supply on the growth of shiitake pure culture.

For the laboratory technician, this study helped determine the best factors affecting the preparation of shiitake pure culture. This study can also serve as basis for individuals going into shiitake mushroom production.

The study was conducted at the Plant Pathology Service Laboratory, Department of Plant Pathology, College of Agriculture, Benguet State University, La Trinidad,

Benguet from November 2005 to February 2006.

REVIEW OF LITERATURE

Classification of Shiitake

The mushroom has been variously placed in the genera *Collybia, Armilloria, Lepiota, Pleurotus,* and *Lentinus*. At this moment, it is now known as *Lentinula edodes*.

According to Stamets (1998), the genus *Lentinula* was originally conceived by Earle in the 1990's and resurrected by Pegler in the 1970's to better define members formerly placed in *Lentinus*. Both genera are characterized by white spores, centrally to eccentrically attached stems, gill edges which are often serrated, and distinct preference for woodland environment. The genus *Lentinula* is monomitic, i.e. lacking dimitic hyphae in the flesh and descending in their arrangement within the gill trama**.** Members in the genus *Lentinus* have flesh composed of dimitic hyphae and highly irregular or interwoven cells in the gill trama.

Description of Shiitake

Stamets (1998) stated that the shiitake mushroom cap measures 5-25 cm broad, hemisphere, expanding to convex and eventually plane at maturity. The cap is dark brown to nearly black at first, becoming lighter brown in age, or upon drying. The cap margin is even to irregular, in rolled at first, then incurved, flattening with maturity and often undulating with age. The gills are white, even at first, becoming serrated or irregular with age, white, bruising brown when damaged. The stem is fibrous, centrally to eccentrically attached, fibrous, and tough in texture. The flesh bruises brown.

Bassett (Undated) reported that the shiitake and other edible mushroom are cultivated on a large scale and complete in commercial importance. Its convex cap varies from white to dark brown and its stem has an upturned ring.

Shiitake Cultivation

Cultivation of this mushroom is a centerpiece of Asian Culture, having employed thousands of people for centuries. The first written record of shiitake cultivation can be traced to Wu Sang Kwuang who was born in China during the Sung Dynasty (960-1127 AD). In 1904, the Japanese researcher Dr. Shozaburo Mimura published the first studies of inoculating logs with cultural mycelium. Once inoculated, logs produce six months to a year later (Stamets, 1998).

Bratkovitch and Baugman (Undated) noted that the log culture, although traditional in Asia, has yet become highly profitable In North America –despite the hopes of many woodlot owners. However, log culture does generate modest supplementary income and fits well within the emerging concept of mycopermaculture .In contrast, Indoor cultivation on sterilized sawdust-based substrates is providing to be highly profitable for those who perfect the technique. Most successful American growers employ methods originating in Asia for the cultivation of this mushroom on sterilized substrates by doubling or tripling the mass of each fruiting block and by “through-spawning”. The Japanese, Taiwanese, and Thai production systems typically utilized cylindrically shaped bags filled with one kilogram of supplemented sawdust, which are top, inoculated. This method gives a maximum of two flushes whereas the more

massive blocks (2-3 kilograms a piece) provide 4 to 5 flushes before expiring.

Przybylowics and Donoghue (1988) noted that the natural method of cultivation could be done on hardwood logs, especially oak, sweet gum, poplar, cottonwood, alder, ironwood, beech, birch, willow, and many other non-aromatic, broad-leaf woods. The denser hardwood produces for as long as six years. The more rapidly decomposing hardwoods have approximately 50% lifespan. The fruitwood is notoriously poor for growing shiitake. Although shiitake naturally occurs on oaks and beeches, the purpose of this mushroom in hardwood stumps in North America had poor success thus far.

MATERIALS AND METHODS

Preparation of Culture Medium

Lentinula edodes, being a basidiomycetous fungus was cultured in potato dextrose agar (PDA). As a factor of the study, PDA was prepared in three ways: as PDA concentrate, as formulated from potato, dextrose and agar, and as formulated from potato, white sugar and gulaman bar. As PDA concentrate, 20 g was stirred continuously in 1000 ml of distilled water at boiling temperature until gelling.

In PDA formulated from potato, dextrose and agar, 200 g of peeled potatoes were boiled in 500 ml distilled water. When the potatoes were cooked, this was filtered and the filtrate was measured. The distilled water was added in the filtrate to make one liter capacity. Afterwards, the 20 g agar and the 20 g dextrose were added into the filtrate. The mixture was heated and continuously stirred while adding the ingredients. When the medium slightly gelled, it was removed from the flame.

In PDA formulated from potato, white sugar and gulaman bar, the 200 g peeled potatoes were boiled in 500 ml distilled water. When the potatoes were cooked, this was filtered and the filtrate was measured and added with distilled water to make one liter. The dextrose was substituted by white sugar at double weight, i.e. 40 g. Likewise the agar was substituted by gulaman bar at double weight. The mixture was heated and was stirred continuously until the gulaman bar and white sugar were fully dissolved.

Preparation of Pure Culture

The healthy mushroom cap was chosen. At the basement of the cap, slices of tissue were cut at about two to three size using a sterilized blade. After which, there

were disinfected with one percent chlorox solution in two to three minutes and washed with 3 series of distilled water. After which, these were blot-dried on sterilized tissue paper to remove excess water on the mushroom tissue. Aseptically, the cut tissues were transferred using a sterilized forcep into the Petri plates containing solidified PDA. Plates were incubated inside the incubation box for about one week until the occurrence of mycelial growth. After one week of incubation, an agar block containing the mycelia was transferred aseptically using sterilized wire loop on PDA slants or desired sterilized containers containing PDA.

Statistical Design and Treatment

The effect of PDA composition as culture medium, and light supply on the growth of shiitake were determined by using the two-factorial completely randomized design (CRD) replicated three times. There were nine treatment combinations with four replicates and three samples per experimental unit. There were a total of 108 culture plates that were evaluated.

The two factors used were the following: the PDA composition as culture medium composition, and light supply. The strain that was used in this study was the BSU selection 505. For factor A (PDA composition as culture medium), these were PDA concentrate, PDA formulated from potato, dextrose and agar and PDA formulated from potato, white sugar and gulaman bar. For Factor B (light supply), these were absence of light, continuous light and alternating 12 hours light and 12 hours darkness. The treatments used were the following:

Factor A = P DA Composition

A_0 = Potato dextrose agar (PDA) concentrate

A_1 = PDA formulated from potato, dextrose and agar

A_2 = PDA formulated from potato, white sugar and gulaman bar

Factor B = Light Supply

B_0 = No light

B_1 = Continuous light

B_2 = Alternating 12 hours light with 12 hours darkness

Data Gathered

The data gathered were:

1. Mycelial growth (mm) at three days after inoculation (DAI). This refers to growth of the mycelium on pure culture measured at three days after inoculation. This was monitored from the point of inoculation to the margin of mycelial growth.
2. Mycelial growth (mm) at 14 days DAI. This was measured as the growth of the mycelium from point of inoculation to the margin of mycelial growth at 14 DAI.
3. Average daily mycelial growth (mm). This refers to the average daily growth of the mycelium.
4. Pure culture quality. This refers to the amount thickness and mature of mycelial growth as observed under the microscope. This was

photodocumented using the micrograph camera.

RESULTS AND DISCUSSION

Mycelial Growth at 3 DAI

Effect of PDA Composition. Table 1 shows that PDA formulated from potato, dextrose and agar gave the highest mean mycelial growth at 3 DAI with 3.24 mm. This was closely followed by PDA concentrate and PDA formulated from potato, white sugar and gulaman bar with mean mycelial growth of 2.25 mm and 1.92 mm, respectively.

Effect of Light Supply. Under light supply, data show that continuous light and alternating 12 hours light with 12 hours darkness gave the best mycelial growth with means of 3.00 mm and 2.67, respectively. Absence of light gave less mean mycelial growth of 1.74 mm.

Interaction between PDA Composition and Light Supply. No significant differences were observed between PDA composition and light supply.

Cost of Production. In terms of production cost of PDA/li., formulation from potato, dextrose, agar (P150) is cheaper than using concentrated PDA (P200). Also, continuous light supply means P10.00 expense and alternation of light and darkness means P4.00 expense.

Mycelial Growth at 14 DAI

Effect of PDA Composition. Concentrated PDA shows the highest mean mycelial growth of 43.42 mm as affected by PDA composition at 14 DAI (Table 2). This was followed by PDA formulated from potato, dextrose, agar (22.08) and PDA formulated from potato, white sugar and gulaman bar (12.33) which did not differ significantly from each other.

Effect of Light Supply. At 14 DAI, light supply did not affect mycelial growth which indicates that *L. edodes* pure culture is not sensitive to light/darkness.

Interaction between PDA Composition and Light Supply. No significant differences were observed between PDA composition and light supply.

Cost of Production. In terms of the cost of producing PDA, the use of concentrated PDA means P200.00 expense which is more expensive than formulating it from potato, dextrose and agar (P150.00) and potato, white sugar and gulaman bar (P50.00). For light supply, continuous lighting means P72.52 expense and alternate light/darkness means P36.30 expense which can be gotten rid off by no light (0 expense) yet equally good mycelial growth.

Average Daily Mycelial Growth

Effect of PDA Composition. Table 3 and Figure 1 shows the average daily mycelial growth as affected by PDA composition and light supply. The concentrated PDA gave the fastest mycelial growth of 3.02 mm. It was closely equally followed by PDA formulated from potato, dextrose and agar and PDA formulated from potato, white sugar and gulaman bar with means of 1.32 mm and 0.72 mm, respectively. It should be noted here that the expensive cost of concentrated PDA gives preference for PDA formulation from potato, dextrose and agar or potato, white sugar and gulaman bar.

Effect of Light Supply. Light supply shows insignificant differences among treatments. Being so, the expenses of continuous lighting and alternation of light and

Table 1. Mean mycelial growth at 3 DAI as affected by PDA composition and light supply

TREATMENT	MEAN MYCELIAL GROWTH(mm)	COST OF PRODUCTION (PhP)
Factor A = PDA Composition		Per Liter PDA
A_0 = PDA concentrate	2.25[b]	200
A_1 = PDA formulated from potato, dextrose and agar	3.24[a]	150
A_2 = PDA formulated from potato, white sugar and gulaman bar	1.92[b]	50
Factor B = Light Supply		Per Kilowatt-Hour
B_0 = No light	1.74[a]	0
B_1 = Continuous light	3.00[a]	14.55
B_2 = Alternating 12 hours light, 12 hours darkness	2.67[a]	7.28

CV = 43.57%

Means in a column followed by a common letter are not significantly different from each other at 5% level DMRT

darkness can be saved by using no light.

Interaction between PDA Composition and Light Supply. No significant differences were observed between PDA composition and light supply.

Table 2. Mean mycelial growth (mm) at 14 DAI as affected by PDA composition and light supply

TREATMENT	MEAN MYCELIAL GROWTH (mm)	COST OF PRODUCTION (PhP)
Factor A = PDA Composition		Per Liter PDA
A_0 = PDA concentrate	43.42[a]	200
A_1 = PDA formulated from potato, dextrose and agar	22.08[b]	150
A_2 = PDA formulated from potato, white sugar and gulaman bar	12.33[b]	50
Factor B = Light Supply		Per Kilowatt-Hour
B_0 = No light	23.25[a]	0
B_1 = Continuous light	30.33[a]	72.62
B_2 =Alternating 12 hours light, 12 hours darkness	24.25[a]	36.31

CV =44.90%

Means in a column followed by a common letter are not significantly different from each other at 5% level DMRT

Cost of Production. In terms of production, the cost of concentrated PDA gave preference for PDA formulation. In light supply, the expenses of continuous lighting and alternation of light and darkness can be saved by using no light.

Table 3. Average daily mycelial growth

TREATMENT	MEAN MYCELIAL GROWTH (mm)	COST OF PDA PRODUCTION (PhP)
Factor A = PDA Composition		Per Liter PDA
A_0 = PDA concentrate	3.02[a]	200
A_1 = PDA formulated from potato, dextrose and agar	1.33[b]	150
A_2 = PDA formulated from potato, white sugar and gulaman bar	0.72[b]	50
Factor B = Light Supply		Per Kilowatt-Hour
B_0 = No light	1.55[a]	0
B_1 = Continuous light	1.98[a]	3.64
B_2 =Alternating 12 hours light, 12 hours darkness	1.53[a]	1.82

CV = 45.82%

Means in a column followed by a common letter are not significantly different from each other at 5% level DMRT

Pure Culture Quality

Effect of PDA Composition. Table 4 outlines the mycelial growth quality observed in the treatments. For concentrated PDA and PDA formulated from potato, dextrose and agar, both had abundant, filamentous white and thick mycelia. On the other hand, PDA formulated from potato, white sugar and gulaman bar had scanty, filamentous, creamy and thin mycelia.

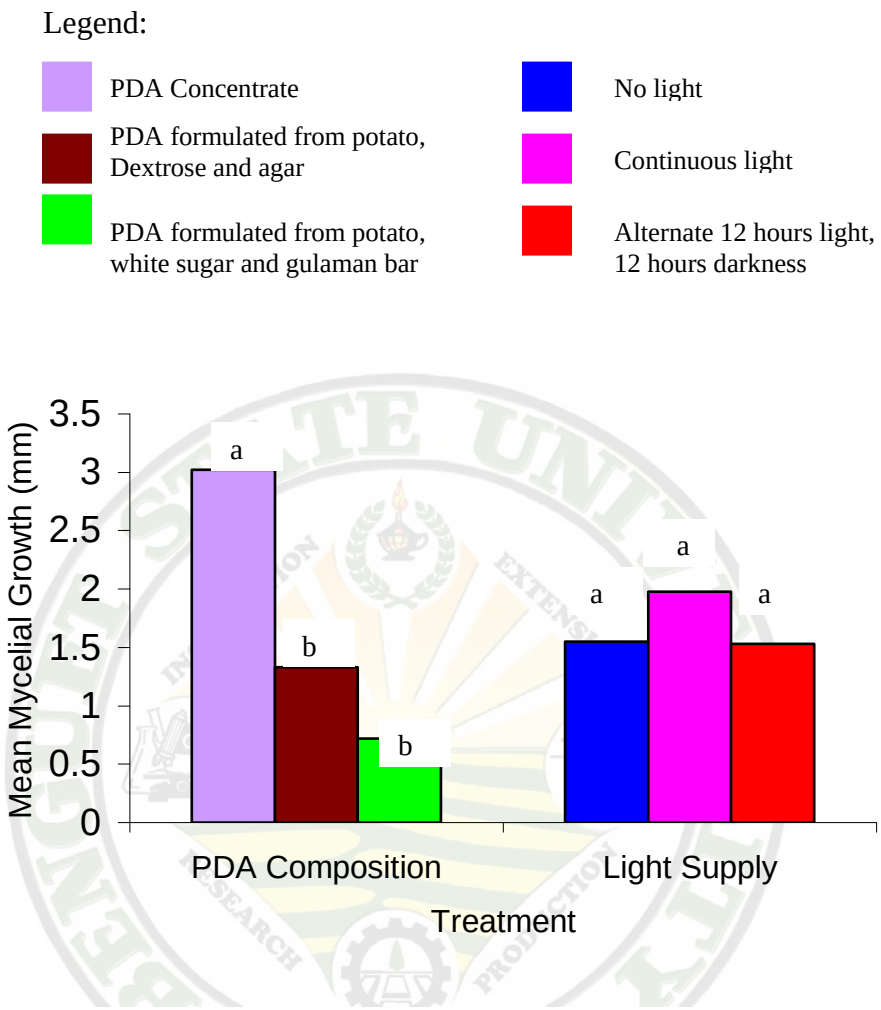


Figure 1. Average daily mycelial growth

Table 4. Pure culture quality of *L. edodes* grown in PDA culture medium with varying ingredients

PARAMETER	PDA CONCENTRATE	PDA FORMULATED FROM POTATO, DEXTROSE AND AGAR	PDA FORMULATED FROM POTATO, WHITE SUGAR AND GULAMAN BAR
Amount of Growth	Abundant	Abundant	Scanty
Form	Filamentous	Filamentous	Filamentous
Color of Mycelium	White	White	Creamy

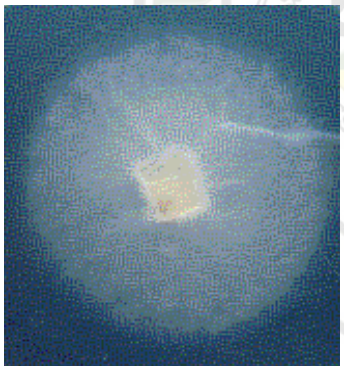
Thickness	Thick	Thick	Thin

PDA Concentrate
No light at 3 DAI

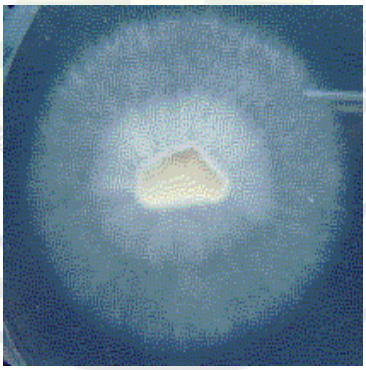
PDA Formulated-
Continuous light
at 3 DAI

PDA Gulaman-
Alternate light (12 hours)
at 3 DAI

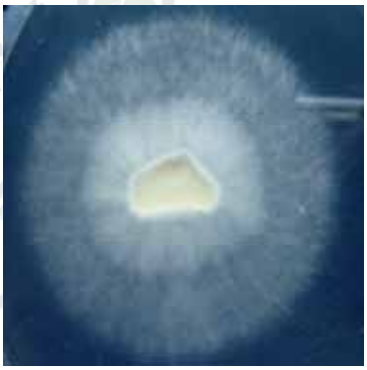
Plate 1. Comparison of *L. edodes* pure culture on PDA composition and light supply at 3 DAI



PDA Formulated-
no light at 14 DAI



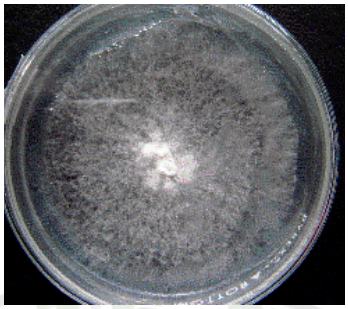
PDA Formulated-
continuous light at
14 DAI



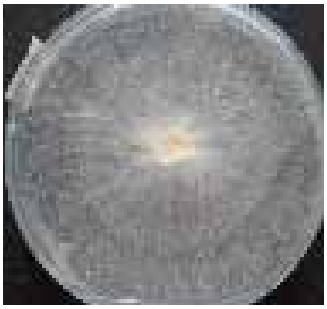
PDA Formulated-
alternate light (12 hours)
at 14 DAI

Plate 2. Comparison of *L. edodes* pure culture on PDA Formulated and light supply at 14 DAI

PDA Concentrate (No light) at 14 DAI

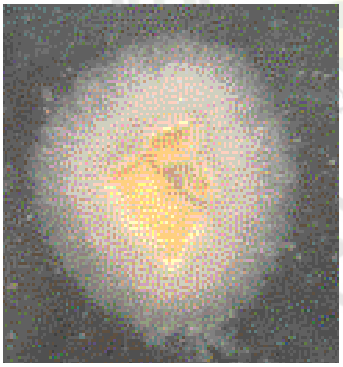


PDA Concentrate (Continuous light) at 14 DAI

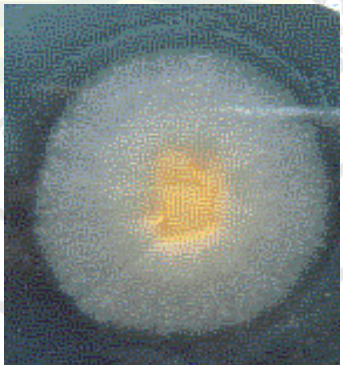


PDA Concentrate Alternate light at 14 DAI

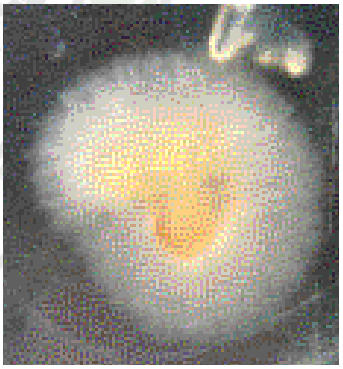
Plate 3. Comparison of PDA Concentrate and light supply at 14 DAI



PDA Gulaman - no light at 14 DAI

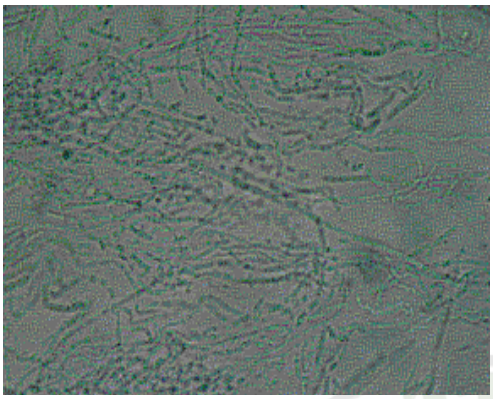


PDA Gulaman - (Continuous light) at 14 DAI



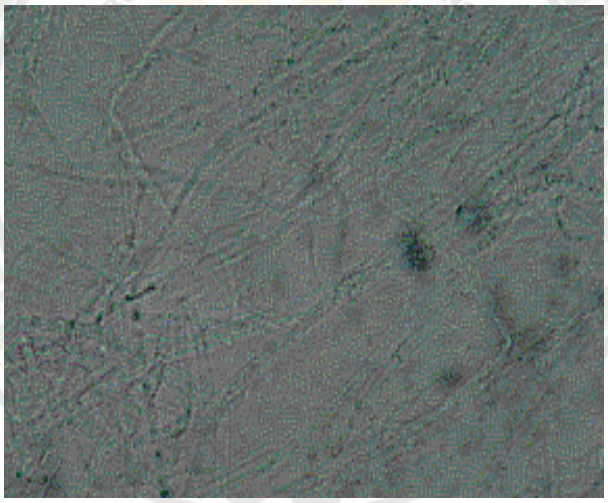
PDA Gulaman - alternate light at 14 DAI

Plate 4. Comparison of PDA Gulaman and light supply at 14 DAI



Microscopic (40x) hyphal observation on PDA Concentrate

Microscopic (40x) hyphal observation on PDA Formulated



Microscopic (40x) hyphal observation on PDA Gulaman

Plate 5. Microscopic (40x) hyphal observations on PDA composition and light supply

SUMMARY, CONCLUSION AND RECOMMENDATION

Summary

The study was conducted to determine the effects of PDA composition as culture medium and light supply on the growth of *Lentinula edodes* pure culture. This was conducted at the Plant Pathology Service Laboratory, Department of Plant Pathology, College of Agriculture, Benguet State University, La Trinidad, Benguet from November 2005 to February 2006.

Based on the results of the experiment, the 3 DAI growth of the mycelia on PDA gave the fastest mean mycelial growth (3.24 mm) and gave better growth when treated with continuous light at 3.00 mm which did not differ significantly from alternating light with darkness at 2.67 mm. The PDA concentrate costs P200 to produce a liter which is more expensive to PDA formulated from potato, dextrose and agar at P150.

At 14 DAI, the highest mean mycelial growth was observed from the PDA concentrate medium with 43.42 mm. Lighting did not affect mycelial growth.

In terms of the average mean daily mycelial growth, PDA concentrate gave the fastest mycelial growth with mean of 3.02 mm. Lighting had no effect on mycelial growth.

In terms of culture quality, concentrate PDA and PDA formulated from potato, dextrose and agar had abundant, filamentous, white and thick mycelia. The PDA formulated from potato, white sugar and gulaman bar had scanty, filamentous, creamy and thin mycelia.

Conclusion

Based on the results, it could be concluded that *L. edodes* mycelia grow fastest at PDA concentrate and does not need light to grow. Considering the cost of PDA concentrate, the PDA formulated from potato, dextrose and agar is an alternative with equally excellent mycelial quality effect.

Recommendation

The pure culture of *L. edodes* is recommended to be isolated in PDA formulated from potato, dextrose and agar and to be incubated under dark condition.

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APPENDICES

APPENDIX TABLE 1. Mean mycelial growth (mm) at 3 DAI as affected by PDA composition and light supply (Actual)

TREATMENT	REPLICATION				TOTAL	MEAN
	I	II	II	IV		
Factor A – Culture Medium Composition						
A_0 - Potato dextrose agar (PDA) concentrate	3.00	2.00	1.67	2.33	9.00	2.25
A_1 – PDA formulated from potato, dextrose and agar	2.33	3.30	3.67	3.67	12.97	3.24
A_2 – PDA formulated from potato, white sugar and gulaman bar	2.00	1.67	1.67	2.33	7.67	1.92
Factor B - Light						
B_0 – No light	1.67	1.30	2.00	2.00	6.97	1.74
B_1 – Continuous light	2.67	2.67	3.00	3.67	12.01	3.00
B_2 – Alternating 12 hours light, 12 hours darkness	3.00	3.00	2.00	2.67	10.67	2.67
Grand Total					59.29	
Grand Mean						2.47

APPENDIX TABLE 2. Mean mycelial growth (mm) at 3 DAI as affected by PDA composition and light supply (Transformed)

TREATMENT	REPLICATION				TOTAL	MEAN
	I	II	II	IV		
Factor A – Culture Medium Composition						
A_0 - Potato dextrose agar (PDA) concentrate	1.87	1.58	1.47	1.68	6.60	1.65
A_1 – PDA formulated from potato, dextrose and agar	1.68	1.45	2.04	2.04	1.71	1.93
A_2 – PDA formulated from potato, white sugar and gulaman bar	1.58	1.47	1.47	1.68	6.20	1.55
Factor B - Light						
B_0 – No light	1.47	1.34	1.58	1.58	5.97	1.49
B_1 – Continuous light	1.78	1.78	1.87	2.04	7.47	1.87
B_2 – Alternating 12 hours light, 12 hours darkness	1.87	1.87	1.58	1.78	7.10	1.77
Grand Total					41.05	
Grand Mean						1.71

ANALYSIS OF VARIANCE

Source of Variance	Degree of Freedom	Sum of Squares	Mean of Square	Computed F	Tabulated F .05	.01
Treatment	11	31.4919	2.863	2.47*	2.22	3.09
Block	3	1.2230	0.408	0.35ns		
A	2	11.4005	5.700	4.92*	3.40	5.61
B	2	10.2005	5.100	4.41*	3.40	5.61
A*B	4	8.6678	2.167	1.87ns		
Error	24	27.7844	1.158			
Total	35	59.2764				

* - significant
ns – not significant

CV = 2.62%

APPENDIX TABLE 3. Mean mycelial growth (mm) at 14 DAI as affected by PDA composition and light supply (Actual)

TREATMENT	REPLICATION				TOTAL	MEAN
	I	II	II	IV		
Factor A – Culture Medium Composition						
A_0 - Potato dextrose agar (PDA) concentrate	50.64	50.00	31.67	41.33	173.67	43.42
A_1 – PDA formulated from potato, dextrose and agar	21.33	22.33	25.67	19.00	88.33	22.08
A_2 – PDA formulated from potato, white sugar and gulaman bar	15.00	9.67	9.67	15.00	49.34	12.33
Factor B - Light						
B_0 – No light	24.00	18.33	25.00	25.67	93.00	23.25
B_1 – Continuous light	35.00	34.67	18.67	33.00	121.34	30.33
B_2 – Alternating 12 hours light, 12 hours darkness	28.00	29.00	23.33	16.67	97.00	24.25
Grand Total					622.68	
Grand Mean						25.94

APPENDIX TABLE 4. Mean mycelial growth (mm) at 14 DAI as affected by PDA composition and light supply (Transformed)

TREATMENT	REPLICATION				TOTAL	MEAN
	I	II	II	IV		
Factor A – Culture Medium Composition						
A_0 - Potato dextrose agar (PDA) concentrate	7.15	7.11	5.67	6.47	26.40	6.60
A_1 – PDA formulated from potato, dextrose and agar	4.67	4.78	5.12	4.92	18.99	4.75
A_2 – PDA formulated from potato, white sugar and gulaman bar	3.94	3.19	3.19	3.94	14.26	3.56
Factor B - Light						
B_0 – No light	4.95	4.34	5.050	5.12	14.46	4.86
B_1 – Continuous light	5.96	5.93	4.38	5.79	22.064	5.52
B_2 – Alternating 12 hours light, 12 hours darkness	5.34	5.43	4.88	4.14	19.79	4.95
Grand Total					120.96	
Grand Mean						5.04

ANALYSIS OF VARIANCE

Source of Variance	Degree of Freedom	Sum of Squares	Mean of Square	Computed F	Tabulated F .05	.01
Treatment	11	7402.389	672.944	4.96^{**}		
Block	3	225.000	75.000	0.55^{ns}	2.22	3.09
A	2	6065.389	3032.694	22.34^{**}		
B	2	352.722	176.361	1.30^{ns}	3.40	5.61
A*B	4	759.278	189.819	1.40^{ns}	3.40	5.61
Error	24	3257.500	135.729			
Total	35	10659.89				

** - highly significant CV = 9.63%

ns – not significant

APPENDIX TABLE 5. Average daily mycelial growth (mm) (Actual)

TREATMENT	REPLICATION				TOTAL	MEAN
	I	II	II	IV		
Factor A – Culture Medium Composition						
A_0 - Potato dextrose agar (PDA) concentrate	3.97	3.50	2.13	2.98	12.08	3.02
A_1 – PDA formulated from potato, dextrose and agar	1.36	1.36	1.48	1.12	5.32	1.33
A_2 – PDA formulated from potato, white sugar and gulaman bar	0.91	0.52	0.57	0.88	2.88	0.72
Factor B - Light						
B_0 – No light	1.60	1.21	1.69	1.71	6.21	1.55
B_1 – Continuous light	2.36	2.33	1.10	2.12	7.91	1.98
B_2 – Alternating 12 hours light, 12 hours darkness	1.78	1.83	1.38	1.14	6.13	1.53
Grand Total					40.53	
Grand Mean						1.69

APPENDIX TABLE 6. Average daily mycelial growth (mm) (Transformed)

TREATMENT	REPLICATION				TOTAL	MEAN
	I	II	II	IV		
Factor A – Culture Medium Composition						
A_0 - Potato dextrose agar (PDA) concentrate	2.11	2.00	1.62	1.87	7.60	1.90
A_1 – PDA formulated from potato, dextrose and agar	1.36	1.36	1.38	1.27	5.37	1.34
A_2 – PDA formulated from potato, white sugar and gulaman bar	1.19	1.00	1.00	1.17	4.36	1.09
Factor B - Light						
B_0 – No light	1.45	1.31	1.48	1.49	5.73	1.43
B_1 – Continuous light	1.69	1.68	1.26	1.62	6.25	1.56
B_2 – Alternating 12 hours light, 12 hours darkness	1.51	1.53	1.37	1.28	5.69	1.42
Grand Total					35.00	
Grand Mean						1.46

ANALYSIS OF VARIANCE

Source of Variance	Degree of Freedom	Sum of Squares	Mean of Square	Computed F	Tabular F .05	Tabular F .01
Treatment	11	41.134	3.739	6.24^{**}		
Block	3	1.356	0.452	0.75^{ns}	2.22	3.09
A	2	34.090	17.045	28.45^{**}		
B	2	1.515	0.758	1.26^{ns}	3.40	5.61
A*B	4	4.172	1.043	1.74^{ns}	3.40	5.61
Error	24	14.379	0.599			
Total	35	55.514				

** - highy significant
ns – not significant

CV = 2.21%