Mycelial Growth Performance of *Pleurotus ostreatus* and *Volvariella volvacea* in Common Kitchen Wastes

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Abstract –*Pleurotus ostreatus (sideways oyster) and Volvariella volvacea (paddy straw) are two of the common mushroom species being cultivated throughout the world that thrives at tropical and sub-tropical countries. The common media used in mushroom cultivation is potato dextrose agar media, which is too expensive for small time mushroom growers. Thus, this study aimed to develop alternative culture media out of kitchen wastes such as Matured Coconut Water (MCW), Rice Wash (RW) and Corn Decoction (CD); specifically it intends to determine the mycelial performance of P. ostreatus and V. volvacea in common kitchen waste media and to evaluate the significant difference of the mycelial growth performance of P. ostreatus and V. volvacea in matured coconut water, rice wash and corn decoction. The pH levels of media were adjusted to 8.0. With 27.47 ^{O}C and 78.83% relative humidity, the fastest mycelial ramification of P. ostreatus was observed on rice wash within six days of incubation which is significantly higher than the two cultured media. However, matured coconut water and corn decoction revealed very thick mycelial density. In addition, all cultured media were fully colonized with P. ostreatus.On the other hand, shortest number of days of mycelial colonization of V. volvacea that took three days was noted in Rice wash (RW) which is significantly different among the evaluated media. In terms of mycelial density, the thickest mycelial ramifications were recorded in coconut water and corn decoction at 28.1 ^{O}C and 79.92% relative humidity. Moreover, all the evaluated media were completely ramified with V. volvacea. These results suggested that rice wash is an ideal media for P. ostreatus and V. volvacea, in addition, matured coconut water and corn decoction can be utilized as an alternative media for P. ostreatus and V. volvacea.*

Keywords – *corn decoction, matured coconut water, mycelial growth, P. ostreatus, rice wash, V. volvacea.*

INTRODUCTION

Mushrooms are ubiquitous in nature. It is heterotrophic organism that depends on dead organic materials for nutrition. Mushrooms are not just functional foods they are also considered as nutriceuticals [1]. Generally, it is consumed because of their good flavour and nutritional value. Moreover, hallucinogens found in mushrooms are utilized by psychiatrist in the treatment of mental illness [2]. There are at least over 1,200 different species of mushrooms reported to be edible. Two of the most cultivated mushrooms in the world are *Pleurotus ostreatus* and *Volvariella volvacea*.

One of the edible mushrooms known to grow on woods is *P. ostreatus* or sideways oyster mushroom, which can be found in tropical and sub-tropical countries [3]. It has broad, fan shaped cap with a diameter of five to twenty-five centimeter [4]. It has a firm white flesh, whitish to cream colored gills and its stipe is attached to woods in a lateral position [5]. Due to simple, low cost production technologies and high biological yield, *P. ostreatus* becomes popular and widely cultivated throughout the world [6]. It has high economical, ecological and medicinal values. Likewise, it can colonize and degrade large variety of lignocellulosic materials and other wastes which are considered as agro-industrial wastes. On the other hand, *V. volvacea* or paddy straw mushroom, is a tropical specie and leaf litter mushroom that thrives best at 35˚C [7]. It has brownish or dark gray colored fruiting bodies [8],and a seven to 10.5 micrometer range of spore size with a length of stipe that exceeds to two centimeter and pileus diameter of five to sixteen centimeter [9]. Because of its pleasant flavour and taste, *V. volvacea* is one of the third most cultivated mushrooms in the world [10]. Thus, demand in healthy food and annual production of *V. volvacea* has increased in recent years [11].

Mushrooms have the ability to transform nutritionally useless wastes into highly acceptable nutritious food [12]. It involves physical and manual labor that creates employment opportunities for laborers and marginal sections of the society. Nowadays, mushroom culture is gaining popularity in the Philippines. Its present cultivation in this country is limited due to limited local knowledge about its cultivation, traditional beliefs of some communities and cultivation of mushroom requires much effort, time and money. However, mushroom production begins from rescuing its healthy cell lines. There are necessary tools, apparatus and media needed in propagating of such fungi. The common media used in mushroom culture is agar media, which is too expensive for small time mushroom growers. However, there are some alternative media that can be use which are proven and less costly. Many researchers have been driven the use of coconut water as an effective media that supports the mycelial growth of different mushroom species [13]. Kitchen wastes such as rice water and corn decoction are wastes from households that emit substances of eutrophication that contributes to environmental problems [14], [15]. Furthermore, the nutritional composition of matured coconut water, rice grains and corn kernels are carbohydrates, proteins and fats [16], [17], [18]. These kitchen wastes can be transformed and develop into an alternative culture media to support the growth of the mushroom mycelia, since it can provide the fungus with the necessary nutrients it needs. Thus, this study aims to determine the mycelial growth of *P. ostreatus* and *V. volvacea* in common kitchen wastes.

OBJECTIVES OF THE STUDY

The objective of this research involved the mycelial growth performance of *P. ostreatus* and *V. volvacea* in indigenous kitchen wastes. Specifically, this study intends to determine the mycelial performance of *P. ostreatus* and *V. volvacea* among matured coconut water media, rice wash and corn decoction in terms of radius and mycelial density; evaluate the significant difference of the mycelial performance of *P. ostreatus* and *V. volvacea* among matured coconut water media, rice wash and corn decoction.

MATERIALS AND METHODS

Research Design

This study used experimental-evaluative method of research to determine the mycelial growth performance of *Pleurotusostreatus* and *Volvariella volvacea* in common kitchen wastes.

Experimental method was used to measure the mycelial growth of *P. ostreatus* and *V. volvacea* in rice wash, corn boiled water and matured coconut water in terms of radius and mycelial density.

Evaluative method was used to determine if there is a significant difference on the mycelial growth performance of *P. ostreatus* and *V. volvacea* among common kitchen wastes media.

Research Setting

This study was conducted at Agro-industrial research building of Central Bicol State University of Agriculture-Sipocot, Zone 5, Impig, Sipocot, Camarines Sur along Maharlika Highway in about 2 kilometers away from Sipocot town proper.The place where the experiment held was in an upland area with a temperature that ranges from 28.3℃ – 27.5℃ and an average relative humidity of 82% - 79.1%.

Research Instrument

The research instruments used in this study are cork borer, ruler and observation sheet. Five millimeter-sized cork borer was used to get a portion of seven-day old mycelial block, ruler (mm) was used to measure the mycelial growth of *P. ostreatus* and *V. volvacea* in prepared medias and observation sheet was used to record the mycelial growth of *P. ostreatus* and *V. volvacea* in common kitchen wastes; it is composed of three treatments (rice wash, corn decoction and matured coconut water media), where in every media, there are three sets of replicates used.

Data Gathering Procedure

Daily mycelial run of *P. ostreatus* and *V. volvacea* were measured at 5:00 pm using ruler (mm) and recorded in observation sheet until the media were fully ramified by mycelia. Temperature and relative humidity were recorded every 7:00 am, 12:00 pm and 5:00 pm.

Experimental Process

This includes the revival of culture, preparation of media, inoculation of test mushroom and incubation of test media.

Revival of culture

Agar blocks are collected from the pure stock culture of *P. ostreatus* and *V. volvacea* from the culture collection of agro-industrial research building of CBSUA-Sipocot.

Preparation of Media

One liter of matured coconut water and one liter of rice wash (1:1 = 1 Kg rice : 1 L water) were added separately with 10 grams of white table sugar and 20 grams of white gulaman bar, boiled until homogenous mixture attained. One kilogram of corn was boiled for 30 minutes in one liter of tap water. After the corn decoction is filtered, 10 grams of white table sugar and 20 grams of white gulaman bar were added and boiled until the solution homogenized. Each solution was volumed up to attained one liter of solution then dispensed in an erlenmeyer flask, plug with cotton and secure with aluminum foil.

Prior to sterilization 0.1 M NaOH or 0.1 HCl, was used to adjust the pH of the media for *P. ostreatus* at pH 8.0, since *Pleurotus* species grew best at this pH [3]; and the favorable pH for mycelial growth of *V. volvacea* is at pH 8.0 [15]. The prepared media for *P. ostreatus* and *V. volvacea* were placed in an autoclave for sterilization process at 15 psi. / 121℃ for 20 minutes.

Inoculation of test mushroom

The newly sterilized media were dispensed in a petri plates and were allowed to cool and solidify. Subsequently, using a five mm cork borer, 7 day old five mm mycelial block was inoculated at the center of the newly solidified media. Each media were sealed with cling wrap.

Incubation of test media

The newly inoculated test media were placed in an incubation chamber under room temperature. The chamber was covered with black plastic since, *P. ostreatus* and *V. volvacea* grows luxuriantly at dark condition [22], [24].

Statistical Treatment

The following statistical tools were used in the study: Weighted mean was used to quantify the mycelial growth performance of *P. ostreatus* and *V. volvacea* in common kitchen wastes. ANOVA was used to determine the significant difference of the mycelial growth performance of *P. ostreatus* and *V. volvacea* among matured coconut water, rice wash and corn decoction.

RESULTS AND DISCUSSION

Mycelial Growth of *P. ostreatus* and *V. volvacea*

Mycelia are vast network of continuously growing filaments of hypha, it is the visible cottony whitish or dirty whitish that spread through the cellulosic material [1]. The mycelial performance of *P. ostreatus and V.volvacea* in matured coconut water (T1), rice wash (T2) and corn decoction (T3) were determined by measuring the daily mycelial run of *P. ostreatus and V.volvacea*.

Presented in table one was the daily mycelial growth performance in millimeter of *P. ostreatus* in common kitchen waste media in terms of radius. On the first day, rice wash elucidated the highest mycelial growth of 2.00 mm while corn decoction showed 1.75 mm which is longer to 0.17 mm of matured coconut water. On the second day, rice wash revealed 8.67 mm and 8.33 mm for corn decoction that was higher to 2.58 mm growth of matured coconut water.

Table 1. Daily Mycelial Growth (mm) of *P. ostreatus* in Common KitchenWastes Media in Terms of Radius

Treatment	Number of days		
	1	**2**	**3**
T1 = Matured Coconut Water	0.17	2.58	8.67
T2 = Rice Wash	2.00	8.67	17.58
T3 = Corn Decoction	1.75	8.33	16.00

Meanwhile, on the third day mycelial run were recorded with 17.58 mm, 16.00 mm for rice wash and corn decoction respectively and 8.67 mm on matured coconut water. Furthermore, *P. ostreatus* fully colonize the rice wash media after six days which is faster than corn decoction that takes seven days to be fully ramified and followed by matured coconut water after eight days of incubation.

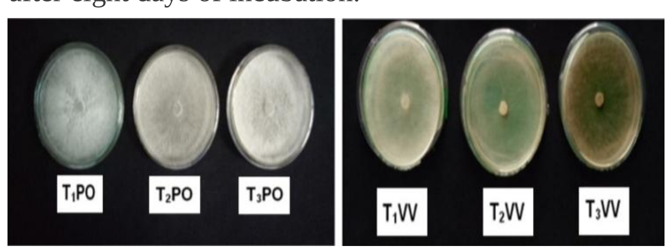

Fig. 1. Mycelial run of *P. ostreatus* after six days and *V. volvacea* after three days of incubation; right (*V. volvacea*) and left (*P. ostreatus*)

The highest growth performance of *P. ostreatus* and *V. volvacea* were recorded in rice wash and corn decoction from first to third day of incubation, it can be attributed to the seven percent protein content of rice wash [30], [37] and six percent corn decoction [38] as compared to matured coconut water with 0.52%. However, this finding was not incongruent with the findings of Zurbano et al. [29], that during 8.2 days of incubation, coconut water gulaman showed the fastest mycelial growth.

Daily growth performance of *V. volvacea* in common kitchen waste in terms of radius was elucidated in table two. On day one in terms of *V. volvacea* mycelial growth, rice wash showed 2.00 mm growth which was longer than corn decoction and matured coconut water with 1.17 mm and 1.67 mm respectively.

On the second day, rice wash revealed 20.42 mm mycelial growth which was longer to 10.58 mm of corn decoction and 12.83 mm of matured coconut water. Additionally, on the third day rice wash fully colonized the media with 42.50 mm mycelial growth which is significantly longer as compared to 28.92 mm of corn decoction and 32.42 mm of matured coconut water.

Table 2. Daily Mycelial Growth (mm) of *V. volvacea* in Common Kitchen Wastes Media in Terms of Radius

Treatment	Number of days		
	1	2	3
T1 = Matured Coconut Water	1.67	12.83	32.42
T2 = Rice Wash	2.00	20.42	42.50
T3 = Corn Decoction	1.17	10.58	28.92

On the first day of observation, the varying mycelial growth of *V. volvacea* in different medias can be attributed to its crude fiber wherein, rice wash, corn decoction and matured coconut water contains thirty, five and three percent respectively. From second to third day the longer mycelial growth of *V. volvacea* in rice wash can be associated to its high content of starch with 75-80%, proteins, iron, phosphorus, potassium, zinc and full complements of amino acid [30], [37].

Table three showed the mycelial growth performance of *P. ostreatus* in different media. In terms of mycelial density, matured coconut water, rice wash and corn decoction elucidated thin mycelial growth on the first, second and third day of incubation.

Table 3. Mycelial Density of *P. ostreatus* on Different Media

Treatment	Number of days		
	1	2	3
T1 = Matured Coconut Water	+	+	+
T2 = Rice Wash	+	+	+
T3 = Corn Decoction	+	+	+

***Note:** The lowest degree marked as + of mycelial thickness, intermediate degree marked as ++, and the highest degree marked as +++ of mycelial thickness [39].

Disclosed in table four was the mycelial growth performance of *V. volvacea* on different media. From first to second day, thin mycelial growth is recorded in matured coconut water, rice wash and corn decoction. However, on the third day matured coconut water and corn decoction showed thick mycelial run while rice wash was noted with thin mycelial growth.

Table 4. Mycelial Density of *V. volvacea* on Different Media

Treatment	Number of days		
	1	2	3
T1 = Matured Coconut Water	+	+	++
T2 = Rice Wash	+	+	+
T3 = Corn Decoction	+	+	++

***Note:** The lowest degree marked as + of mycelial thickness, intermediate degree marked as ++, and the highest degree marked as +++ of mycelial thickness [39].

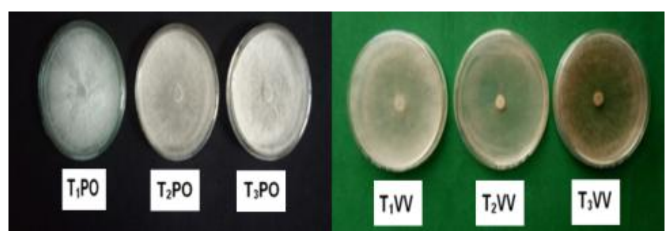

Figure 2. Mycelial density of *P. ostreatus* after six day and *V. volvacea* after three days of incubation; left (*P. ostreatus*) and (*V. volvacea*)

Difference on the Mycelial Growth of *P. ostreatus* and *V. volvacea* among Matured Coconut Water, Rice wash and Corn Decoction

Mycelial growth performance of *P. ostreatus* and *V. volvacea* in common kitchen wastes medias were analyzed and interpreted through weighted mean and Anova.

Table five presents the significant difference on the mycelial growth performance of *P. ostreatus* on different kitchen wastes media after three days of incubation. On the first to third day of observation, data revealed that matured coconut water, rice wash and corn decoction revealed no significant difference, since the F-test value on the first day was 0.08, 0.03 for the second day of observation and 0.02 on the third day of incubation and were found lesser than the critical test value of 5.14.

Table 5. Difference on the Mycelial Growth of *P. ostreatus* on Different Kitchen Wastes Medias After Three Days of Incubation

No. of Days	Tc	Level of Significance	Critical Test Value	Decision	Interpretation
1	0.08	0.05	±5.14	Accept H_o	Not Significant
2	0.03				Not Significant
3	0.02				Not Significant

Since the computed value of the number of days of incubation of *P.ostreatus* and *V. volvacea* in different kitchen wastes media were found lesser than the critical value, the null hypothesis was accepted that there is no significant difference on the mycelial growth of *P. ostreatus* on different kitchen wastes. Thus, these finding suggested that Matured coconut water, rice wash and corn decoction can be used as an alternative media.

As recorded on table six, mycelial growth performance of *V. volvacea* on matured coconut water, rice wash and corn decoction were not significantly different, based from the computed value of 1.06, 0.03 and 0.03 for the first to third day of observation respectively which were less than the critical test value of 5.14.

Table 6. Difference on the Mycelial Growth of *V. volvacea* on Different Kitchen Wastes Medias after Three Days of Incubation

No. of Days	Tc	Level of Significance	Critical Test Value	Decision	Interpretation
1	0.08	0.05	±5.14	Accept H_o	Not Significant
2	0.03				Not Significant
3	0.02				Not Significant

Table 6 elucidated that null hypothesis was accepted that there is no significant difference on the mycelial growth performance of *V. volvacea* on different kitchen wastes, since the computed value of the number of days of incubation of *V. volvacea* in different kitchen wastes media were found lesser than the critical value. Thus, these finding suggested that matured coconut water, rice wash and corn decoction can be used as an alternative media in culturing *V. volvacea*.

CONCLUSIONS

Within six days of incubation, rice wash generated the highest mycelial growth of *P. ostreatus* than the matured coconut water and corn decoction kitchen waste media. Corn decoction is the second treatment that produced high mycelial growth and completely colonized the media within seven days. Matured coconut water produced the slowest mycelial growth performance and ramified the media within eight days. Furthermore, Data showed that within six days of incubation, matured coconut water and corn decoction produced the thickest mycelial growth while rice wash generated thin mycelial run.

Data revealed that within three days of incubation period, rice wash produced the longest and fastest day of mycelial ramification of *V. volvacea* in three days. However, the slowest mycelial growth is noted on corn decoction that ramified the media within four days together with coconut water. Moreover, findings revealed that rice wash generated thin mycelial growth of *V. volvacea*. Furthermore, after three days of observation matured coconut water and corn decoction showed thick mycelial growth.

Common kitchen wastes such as rice wash and corn decoction can be compared to matured coconut water media since they have the ability to support the mycelial growth of *P. ostreatus* and *V. volvacea*.

RECOMMENDATIONS

Rice wash is ideal media for *P. ostreatus* and *V. volvacea*, however, all the evaluated media; matured coconut water, rice wash and corn decoction can be utilized as culture media, since it was fully colonized with mycelia.

Other kitchen wastes could be evaluated as alternative media for mycelial production of *P. ostreatus* and *V. volvacea*.

Corn decoction and matured coconut water can be an alternative media since it has an even luxuriant mycelial run and consider other physical factors that might affect the ramification, incubation and proliferation of the mycelia such as the nutrient content, pH, temperature, relative humidity, lighting condition and the agents of contamination.

Appendixes

Difference on the Mycelial Growth of *P. ostreatus* on Different Kitchen Wastes Medias (Day 1)

F-test value= $\frac{S^2_W}{S^2_B}$
F-test value= $\frac{0.20}{2.97}$
F-test value= 0.08
Critical Test Value = ±5.14
Decision: Accept Ho
Interpretation: There is no significant difference

Difference on the Mycelial Growth of *P. ostreatus* on Different Kitchen Wastes Medias (Day 2)

F-test value= $\frac{S^2_W}{S^2_B}$
F-test value= $\frac{0.96}{35.13}$
F-test value= 0.03
Critical Test Value = ±5.14
Decision: Accept Ho
Interpretation: There is no significant difference

Difference on the Mycelial Growth of *P. ostreatus* on Different Kitchen Wastes Medias (Day 3)

F-test value= $\frac{S^2_W}{S^2_B}$
F-test value= $\frac{1.34}{66.96}$
F-test value= 0.02
Critical Test Value = ±5.14
Decision: Accept Ho
Interpretation: There is no significant difference

Difference on the Mycelial Growth of *V. volvacea* on Different Kitchen Wastes Medias (Day 1)

F-test value= $\frac{S^2_W}{S^2_B}$
F-test value= $\frac{0.55}{0.52}$
F-test value= 1.06
Critical Test Value = ±5.14
Decision: Accept Ho
Interpretation: There is no significant difference

Difference on the Mycelial Growth of *V. volvacea* on Different Kitchen Wastes Medias (Day 2)

F-test value= $\frac{S^2_W}{S^2_B}$
F-test value= $\frac{2.21}{75.02}$
F-test value= 0.03
Critical Test Value = ±5.14
Decision: Accept Ho
Interpretation: There is no significant difference

Difference on the Mycelial Growth of *V. volvacea* on Different Kitchen Wastes Medias (Day 2)

F-test value= $\frac{S^2_W}{S^2_B}$
F-test value= $\frac{5.03}{149.14}$
F-test value= 0.03
Critical Test Value = ±5.14
Decision: Accept Ho
Interpretation: There is no significant difference

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