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Mycelial Growth with Nutrient Supplementation in Pure Culture and Yield, N-Analysis and Income Potential of Paddy Straw Mushroom (*Volvariella volvacea*) With Different Substrates

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ABSTRACT

The insufficient planting materials and lack of technical know-how of *V. volvacea* cultivation is the one of the major problem on technology transfer for mushroom production. This research study was conducted to validate and evaluate the performance of *Volvariella volvacea* with different nutrient sources and substrates. This study was composed of two-parts, mycelial growth performance with different growing media (Study 1) and yield, N-analysis and income potential (Study 2). Study 1 was a single-factor experiment conducted in a randomized complete block design (RCBD) and Study 2 used 3x5 factorial experiment also in RCBD. Test for significant differences among treatment means was done analysis of variance (ANOVA) and pairwise comparison was done with Least Significant Difference test (LSD). Analysis was facilitated by the use of statistical software. Sweet potato has the higher potential as growing media for pure culture of *V. volvacea* fungal inoculation. Some characteristics such as pinhead size, stipe length and biological efficiency were affected by nutrient supplement and kind of substrate. However, the most important variable which is yield was affected by substrate only. Regardless of the nutrient sources *V. volvacea* will grow under different substrates such as banana leaves, corn bagasse, water hyacinth and rice straw except wood shavings. Total nitrogen content converted into protein (%) was undertaken to determine its component for various purposes. Biological efficiency of substrates was higher with banana leaves, rice straw and water hyacinth is significantly different as it is supplemented with inorganic fertilizer (urea) or no nutrient added. Mushroom production under different substrates supplemented with inorganic fertilizer can give promising income. Further research and exploration on the commodity may be conducted to ensure higher efficiency in production that can be used for community acceptability, thus, contribute to increasing the supply of mushroom. As a viable enterprise, mushroom production can be engaged by farmers and households for socioeconomic upliftment.

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KEYWORDS: V. volvacea, locally available substrates, nutrient sources, mycelial growth, supplementation.

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INTRODUCTION

Mushroom is a fleshy saprophyte fungus commonly found growing on rotten log of wood trunk of trees, decaying organic matter and in damp soil rich in organic substances. Edible mushroom is highly nutritious and can be compared with eggs, milk and meat (Oei, 2003). The content of essential amino acids in mushroom is high and close to the need of the human body that which is easily digestible and has no cholesterol content. However, the cultivation of mushroom is still limited, and the industry is still at its infancy (Belewu, 2002 and 2003).

Commonly, straw mushrooms are grown on rice straw beds and are most commonly picked when immature often labeled "unpeeled" during button or egg phase and before the veil ruptures (Chang et.al.,1982). Straw mushrooms are adaptable to its environment and takes four to five days to mature. Mostly, it successfully grown in subtropical climates with high annual rainfall.

With its use and fucntions, Hsiung (2006) pointed out that there were no record of straw mushroom cultivation before the 19th century.

The brief period of wild mushroom germination coincides with the arrival of the storm or rainy season in the Philippines. Strong lightning, which typically happens in the rainy season, raises the concentration of nitrate compounds in the atmosphere, which in turn causes wild mushrooms to sprout along farms and forests, especially on anthills that are decomposing and other natural areas with decomposing organic materials like leaves, wood, and animal dung. Thus, in order to meet market demand, all varieties of mushroom output must be increased.

The current total mushroom production of the Philippines is 463 mt (BAS 2014) regardless of variety. Among the 17 regions, CALABARZON, Central Luzon, Davao, Cagayan Valley and Ilocos are the regions with the greatest contributions in mushroom production. Mushroom importation totaled to 656,253 kg with a total value of US$343,179 (BAS 2014). Imported mushroom varieties are mostly specialty mushroom varieties such as Shiitake, white button mushrooms, winter mushrooms, earwood mushroom and king oyster mushrooms. According to the Department of Agriculture's most recent data (2021), mushroom production is predicted to increase from 689 metric tons in 2021 to 743 metric tons in 2026, a 1.3% annual rise. The nation has had an annual growth rate of 3.5% since 1987.

According to Chang et al. (2014), Philippines experience tropical climate where the condition is the best for mushroom cultivation. Considering the situation, it will be a turning point for growers to try growing mushroom. In addition, the Philippine government started a 5-year plan for mushroom research and development initiatives. According to the idea, mushroom output might rise, which would enhance the living conditions for the farmers. Furthermore, the history of mushroom production in the Philippines is really concise because in its succeeding years the production has increased as day passed by. Mushroom production is about 700 tons to 800 per year until year of 1997 but it declined to 500 tons to 600 tons per year during 1998 to 2008. Unfortunately, there's a sharp reduction in the production in 2009 as it had 300 tons to 400 tons per year. Today, the level of self-production for mushroom is only 5%. Thus, the Department of Agriculture is putting up Mushroom Technology Center in Tarlac which costs Php8 million to boost the production of mushroom and prevent the country from importing. The Department of Agriculture (2021) also states in the said journal that there was an unpredictable production of mushroom occurred because of growers of mushroom became interested on rice as the government emphasizes rice industry by boosting different programs. However, since the production of mushroom achieve its minimum level during 2009, the output shave been still increasing every year with the help from Department of Agriculture and other local universities. The mushroom industry in the Philippines is manage by many small business or producers. Yet, there are still large-scale producers located at Tagaytay, Batangas and Baguio City. Despite of this, Philippines still imports around 150 metric tons of mushroom yearly in Taiwan, China, Thailand and Japan (Chang et al., 2014).

The mushroom industry is profitable, according to Medenilla (2020), and local mushroom growers split the profits. However, the Philippines only contributes 10% of the local mushroom demand to date, largely because many Filipinos are not aware of the business opportunities in mushroom farming. Additionally, the mushroom industry just requires hard work and dedication to succeed; it provides consumers with a healthy option as well as prospects for money and living for potential farmers.

As consumers' preferences for organic food grow, developing economies are predicted to expand across the board. The cost of mushrooms is lower in nations like South Korea, India, and others where specific varieties, like button mushrooms, are grown in large quantities. This could boost sales in the aforementioned nations because, thanks to innovation, mushrooms can now be used in place of many food products (Fortune Business Insights, 2019). The COVID-19 pandemic has negatively impacted the growth of the global mushroom business. Travel restrictions brought about by lockdowns around the world have had a significant impact on the market dispersion channel design of the emerging mushroom business. Additionally, because of the weak market demand, manufacturing facilities have ceased operations, which has resulted in enormous losses for the owners of mushroom farms, who are solely dependent on the agricultural industry. In the meantime, the health crises create a new avenue for the promotion and supply of mushrooms through online social media platforms, reaching a wider audience than with conventional marketing and distribution methods (Kadam & Deshmukh, 2021).

A type of edible fungus called paddy straw mushroom (*V. volvacea*) is widely utilized in Asian cuisines and is grown throughout East and Southeast Asia. In the areas where they are grown, they are usually found fresh; in other places, they are more commonly found dried or tinned. Straw mushrooms rank third in the world's mushroom consumption.

In the Philippines, mushroom production has been established way back in the 19th century. Considering that mushroom had been long introduced by various government and non-government institutions, the Philippines contribution to the local mushroom demand is only at 10% to date (Medenilla, 2020).

Although it can be a profitable business, many Filipinos do not venture into mushroom farming due to lack of awareness. However, cultivating mushrooms can provide one with a sustainable income as supplemental source of income for a farmer. In the Philippines, it is a common practice of burning straw after harvest of rice. These agro-waste materials can be useful in mushroom production even in a paddy field as a growing medium. Instead of burning the rice straw, it can

be used to produce mushrooms that will not only save the environment through the lessening of agricultural wastes (like rice straw, dried banana leaves, rice hull, and sawdust), but it can also help earn more.

In the Philippines, mushroom culture is becoming more and more popular. A delicacy that is truly recognized as a vegetable is the mushroom. The food in question is considered useful and has been identified by the industry as a feasible means of creating more consistent revenue streams. This is particularly true for small-scale farmers, as it requires minimal acreage, a short growing season, and low production costs. The Philippines is a very viable place to produce mushrooms since there are plenty of inexpensive raw materials, low production costs, and potential mushroom growers. V. volvacea mushrooms have the ability to be cultured and produced in accordance with the common mushrooms that are now cultivated, thanks to a climate that is suitable to the growth and development of mushrooms (Medenilla, 2020). However, the cultivation of mushroom is still very limited and the industry is still at its infancy to sustain the increasing demand of this commodity.

Based on the Harmonized Research and Development Agenda 2017-2022 and 2022-2028 of the Department of Science and Technology, mushroom was listed as one of the priority commodities for research and development undertakings. Mushroom is one of the high value crops listed by the Department of Agriculture (2022) that should be transferred to the farmers for production and income. Moreover, DOST (2022) pointed out that the RDE for mushroom is focused on production technology, post-harvest technology among others. Also, the agriculture Research Development and Extension sector supports organic agriculture, halal food production, food safety and traceability initiatives, and the development of genetically modified organisms as long as it is compliant with biosafety rules and regulations (DA, 2022).

Present cultivation in this country is limited, perhaps due to insufficiency of planting materials and the limited local knowledge about its culture. The absence of technological know-how for growing mushrooms is the main issue with the transfer of technology for this purpose. In this study, introduction, cultivation and utilization of agricultural wastes are used to evaluate technologies that would be extended to the community to help not only the farmers but the general public to use their vacant areas in mushroom cultivation and hence generate income.

OBJECTIVES

Generally, the study aimed to assess and evaluate the performance of *V. volvacea* with different nutrient sources and substrates in its cultivation and production. Specifically, the study:

1. Determined the mycelial growth performance of V. volvacea with different growing media such as rice wash, matured coconut water and sweet potato decoction, in terms of daily mycelial run (mm).
2. Determined the effect of different nutrient supplements on the performance of V. volvacea in two experimental locations terms of (a) pinhead formation (days) and (b) Growth and yield
3. Identified the total nitrogen content of V. volvacea grown under different substrates supplemented with various nutrient sources.
4. Determined the biological efficiency of the substrates used.
5. Identified the economic value of Volvariella mushroom in terms production income and cost analysis.
6. Determined the significant differences on the mycelial growth performance of V. volvacea with different growing media such as rice wash, matured coconut water and sweet potato decoction, in terms of daily mycelial run (mm);
7. Determined the significant differences on the effect of different nutrient supplements on the performance of V. volvacea in two experimental locations terms of (a) Mycelial emergence to Pinhead formation (days) and (b) Growth and yield.

METHODOLOGY

The study was consist of studies that focused on Mycelial Growth of *V. Volvacea* in Pure Culture with Different Growing Media and Yield and proximate analysis of *V. Volvacea* grown in different substrates and nutrient supplements.

Research Design

Study 1. Mycelial Growth of V. Volvacea in Pure Culture with Different Growing Media

This study determined the mycelial performance of *V. volvacea* using common kitchen waste such as matured coconut water, sweet potato concoction and rice wash base for agar. This study utilized experimental-evaluative method of research to determine the mycelial growth performance of *V. volvacea* in common kitchen wastes. Complete block design was used for the distribution of treatments. Four (4) petri-dish was used that corresponds to replication per growing media to substantiate the result of the experiment. The distribution of treatments was done randomly using the Picker Wheel, an online application used to decide a random choice per blocks.

Study 2. Yield and proximate analysis of V. Volvacea grown in different substrates and nutrient supplements

A 3x5 factorial experiment following a Randomized Complete Block Design (RCBD) with 5 replications was used in this study. The experiment utilized the following factors. The levels of Factor A (nutrient supplement) and Factor B (substrates material) are shown in Table 1. Table 2 shows the factor combinations and code used for each combination.

Table 1. The materials for nutrient supplement and substrate

Factor A. Nutrient supplement	Factor B. Substrate Material
O – Control	RS – Rice Straw (shredded)
U – Urea	WS – Wood shavings
V – Vermicast	CB – Corn bagasse (shredded)
	WH – Water hyacinth(shredded)
	BL – Banana Leaves (shredded)

Table 2. Combination of nutrient supplement and substrate

Factor Combination	Code
No nutrient supplement, Rice Straw	0-RW
No nutrient supplement, Wood shavings	0-WS
No nutrient supplement, Corn bagasse	0-CB
No nutrient supplement, Water hyacinth	0-WH
No nutrient supplement, Banana leaves	0-BL
Urea, Rice Straw	U-RW
Urea, Wood shavings	U-WS
Urea, Corn bagasse	U-CB
Urea, Water hyacinth	U-WH
Urea, Banana leaves	U-BL
Vermicast, Rice Straw	V-RW
Vermicast, Wood shavings	V-WS
Vermicast, Corn bagasse	V-CB
Vermicast, Water hyacinth	V-WH
Vermicast, Banana leaves	V-BL

The distribution of the factor combinations as treatments in each block was done randomly using the Picker Wheel, an online application used to decide a random choice per block. All information on Picker Wheel is protected by copyrights, trademarks, and/or other intellectual property rights that are either owned and controlled by Picker Wheel or by third parties who have granted licenses or otherwise contributed their content to the website. This includes, without limitation, names, logos, trademarks, images, text, columns, graphics, sound effects, musics, videos, photographs, illustrations, artwork, software, and other elements.

Subjects of the Study

The V. volvacea Mushroom

The subject of the study was the paddy straw mushroom (*Volvariella volvacea*). *V. volvacea* commonly known as "warm mushroom" as it grows at relatively high temperature. For this mushroom to grow, the ideal temperature and relative humidity are 30 to 35°C and 80 to 90%, respectively. It is a mushroom that grows quickly; in ideal growth circumstances, the entire crop cycle can be finished in three to four weeks (Royse, 2007).

The Growing Medium

Also, subjected in this study are the matured coconut water, rice wash and sweet potato decoction as growing medium for pure culture. Coconut water was extracted to a fully matured coconut purchased from the farmers at Siniloan Public Market, Siniloan, Laguna. It was purchased without husk and no damage on its coconut shell.

Moreover, rice wash used in the study was came from one (1) kilogram Rc 216 rice harvested at DOST DAT-BED project. Further, sweet potato used in the study was purchased at Siniloan Public Market prior to the conduct of the study. Sweet potato used is classified as medium size with approximate length of five (5) inches.

The Nutrient Supplements

In terms of nutrient supplementation sources - urea and vermicast were used. The inorganic nutrient source was bought in a reputable agricultural supply store in Siniloan, Laguna while the organic nutrient source came from LSPU Mushroom Research and Production Center. The vermicast has N-content of about 3.21%N based on the analysis of samples submitted by Balita (2020) using different nitrogen enriched substrates.

The Substrates

Likewise, different substrate materials locally available in the experimental location was subjected to test in this study. This includes rice straw, wood shavings with rice bran, corn baggase, water hyacinth and banana leaves.

Rice straw was collected from LSPU DOST-DATBED Rice Project three (3) days of harvesting. Collected rice straw was piled in Men's Dorm Building located in the university to undergo pre-decomposition. There were total of eight (8) sacks of rice straw was collected. A 150mL hydrogen peroxide diluted with 16L of water was used to disinfect the piled rice straw.

On the other hand, wood shavings was collected from a furniture shop located at Matalatala, Mabitac, Laguna. The wood shavings was collected from the bottom portion of piled shavings in the disposal area. This is due to the purpose that the wood shaving is pre-decomposed due to the presence of moisture. The color of wood shavings was reddish and identified by the shop owner as shavings from mahogany woods which primarily their source of raw materials.

Moreover, corn bagasse was purchased at Lumban, Laguna from a corn farmer engaged in the industry for 12 years. A 10-inch cut size of corn bagasse was stocked in a sack and transported to LSPU Siniloan for disinfection using 150mL hydrogen peroxide diluted to 16L of water.

Further, water hyacinth was collected along LSPU College of Agriculture water drainage and nearby rice fields. It was collected for the purpose of this study and as well to clean up the drainage canals and rice field for its production. It was collected fresh and sun dried for 17 hours to remove moisture. Furthermore, dried banana leaves were collected from a banana plantation located at Brgy. Banilan, Pakil, Laguna. Collected banana leaves were piled at a storage nearby the house of the owner and transported to LSPU for disinfection using 150mL hydrogen peroxide diluted in 16L of water.

Sampling Technique

For Study 1, four (4) petri dishes were used as sample from each of the growing medium used. This was used for measurement of mycelial growth.

On the other hand, Study 2 utilized 75 styro boxes that consists of 15 combinations each replicated 5 times. On the other hand, a farmer cooperator who has engagement in mushroom production was tapped to undertake the experiment. Experimental set up for Study 2 was followed. Complete enumeration of mushroom bodies for measurement of pin head and stipe was done with the harvested mushroom.

For the yield component, all harvested mushroom was weighed using digital weighing scale.

For analysis on total nitrogen, Kjeldhal distillation method was undertaken. Fifteen mushroom samples with 10 grams per combination from LSPU set-up were randomly selected from five blocks for laboratory analysis. Fresh *V. volvacea tissue* was placed in laundry net and sun dried. The tissues used for nitrogen analysis was the cap, stipe and the volva.

Data Gathering Procedure

Collection of data. For Study 1, daily mycelial run of *V. volvacea* was measured at 5:00 pm using ruler (in mm) and recorded in observation sheet until the media was fully ramified by mycelia which happened in four days.

For Study 2, all 75 samples were subjected to data collection. In this study, the data collection was undertaken daily every 7:00 o'clock in the morning, 12:00 noon, 3:00 o'clock and 5:00 o'clock in the afternoon. However, there are instances that when there is available mushroom even in nighttime, samples were collected. From first harvest samples were collected and placed into plastic tray for measuring of stipe length, pin head and weight. This was basis in computing the marketable weight and biological efficiency of *V. volvacea.* All needed data were collected and recorded in a record book.

Complete enumeration for mushroom bodies for measurement of pin head and stipe was done with the harvested mushroom. Pinheads of *V. volvacea* was measured using caliper. The stipe of mushroom was measured from the tip of the head and the base with the use of ruler.

The yield and yield attributing parameters for example, days taken for spawn run, pin head formation, time taken at first harvest was recorded in each treatment. The biological efficiency was calculated using the following formula.

$$\text{Biological Efficiency (\%)} = \frac{\text{Fresh weight (g) of mushroom harvested}}{\text{Dry weight (g) of substrate}} \text{ x100}$$

Nitrogen content of *V. Volvacea* particularly the total N of mushroom samples was submitted for laboratory test using Kjeldahl distillation method at the Institute of Chemistry - Analytical Services Laboratory, University of the Philippines at Los Baños.

Microclimatic factor data in the experimental area such as minimum temperature (Tmin), maximum temperature (Tmax), and relative humidity (RH), were gathered. The average temperatures were presented from April 2022 to August 2022 when the study was conducted. Due to circumstances that the sample was contaminated, and set-up was repeated, the data on climatic condition was gathered using the mobile application in a cellular phone inside the experimental set-up. The application was Thermometer Room Temperature developed by ZM Technologies installed in Realme C25s. The application was calibrated during the period that the data were collected.

Data Processing and Statistical Procedure

Data gathered were examined utilizing the Analysis of Variance (ANOVA) in Completely Randomized Design (CRD) for Study 1 and was used to describe the mycelial growth performance and yield of *V.* volvacea. For testing of significant differences among treatment means and interaction effects, ANOVA in Randomized Complete Block Design (RCBD) with combined analysis using two locations was used to determine the significant differences among the treatments means in Study 2. Treatment means were compared using Least Significant Differences (LSD). To facilitate easy computation, the data were analyzed using Microsoft Excel and Statistical Tool for Agricultural Research (STAR).

RESULTS AND DISCUSSION

Climatic Data

Table 3 shows the average monthly climatic data. Data show that the highest average monthly temperature was noted on the month of May 2022 in Siniloan and Lumban, Laguna with average temperature of 29.71℃ and 28.84℃, respectively. On the other hand, the highest relative humidity (RH) (87.56%) was observed in the month of August 2022 in Lumban and 83.75% in Siniloan.

Table 3. Monthly maximum, (Tmax), minimum (Tmin), and mean temperatures (Tmean) in -°C, and RH in % at the two experimental areas.

Month	Climatic Data							
	Siniloan, Laguna				Lumban, Laguna			
	Tmax	Tmin	Ave.	RH	Tmax	Tmin	Ave	RH
April	33.06	24.65	28.85	70.64	32.35	25.23	28.79	69.12
May	34.14	25.29	29.71	86.97	31.86	25.83	28.84	75.45
June	33.41	20.79	27.10	76.12	30.19	25.31	27.75	82.38
July	32.62	24.75	28.68	80.77	28.92	24.93	26.92	85.18
August	31.36	21.22	26.29	83.75	28.71	24.85	26.78	87.46
Mean	32.92	23.34	28.13	79.65	30.41	25.23	27.82	79.92

Attempts to cultivate the *V.volvacea* mushrooms with biotechnologies have not yielded satisfactory results until about 2000. The native tropical medium of *V.volvacea* is very difficult to reproduce in culture (Ellertsen, 2005). *V. volvacea* mushroom, has high temperature, light and humidity requirements (Park 2001, Chen 2003, Siwulski and Sobieralski 2004). According to Chang (2008), mycelium of this species grows over a relatively wide range of temperatures from 15°C to 35°C, while Colauto et al. (2008) mentions temperatures between 22°C and 34°C.

In contrast, optimal temperature data are relatively divergent and fall within the following values: 28-31 °C (Colauto et al., 2008), 28-30 °C (Neves et al., 2005), 25-30 °C (Siwulski and Sobieralski 2004), 25-28 °C (Mendonca et al. 2005), 23-27 °C (Chang 2008) and 20-33 °C (Huang 1997). During the incubation period of the mycelium, in the first stage of cultivation, the temperature

should be between 23 and 27 °C (Huang 1997, Iwade and Mizuno 1997). In turn, Stamets (2000) recommended a range of slightly wider temperature from 21 to 27° C. Mendonca et al. (2005) noted that during the period of mycelium growth, the temperature should be kept within the range of 25-30 °C. The monthly temperature and relative humidity that prevailed in the experimental conditions of the present study were within those observed or recommended in the literatures cited above.

Mycelial growth performance of *V. volvacea* with different growing media in pure culture

The data on cumulative daily mycelial run under different growing media in pure culture are presented in Table 4. At the last day of observation (Day 4), the length of mycelia were 28.38mm in coconut water, 21.00mm in rice wash, and 30.50mm in sweet potato decoction. In sweet potato decoction, there was a decrease in mycelial growth at Day 2 but a sharp increase at Day 3. For coconut water, daily mycelial increase was observed until Day 4.

Significant differences in the mean mycelial run rates among treatment means for Days 1-4 were revealed by analysis of variance. Pairwise comparison showed that at Day 1, sweet potato decoction had a mean significantly higher than those of matured coconut water and rice wash. On Day 2, matured coconut water had significantly greater mycelial run compared with rice wash and sweet potato decoction. On Days 3 and 4, mycelial growth rate was same for matured coconut water and sweet potato decoction and both were significantly higher than that of rice wash.

Table 4. Mean daily cumulative mycelial run rate (mm) of *V. volvacea* under different growing media.

Growing Media	Mycelial Run (mm)			
	Day 1	Day 2	Day 3	Day 4
Coconut water	12.25^{b}	20.25^{a}	24.75^{a}	28.38^{a}
Rice wash	10.75^{b}	10.75^{b}	15.75^{b}	21.00^{b}
Sweet Potato	15.25^{a}	12.50^{b}	27.62^{a}	30.50^{a}
Mean	12.75	14.50	22.71	26.62

Analysis of variance

Source	CV	F value	P-value
Growing Media (Day 1)	14.38	6.25	0.019*
Growing Media (Day 2)	8.13	73.62	0.000**
Growing Media (Day 3)	10.19	19.15	0.000**
Growing Media (Day 4)	5.27	50.60	0.000**

The results of analysis show that the material used as pure culture media significantly affect the growth of *V. volvacea* mycelia. Mixture of coconut water has been extensively used as a growth-promoting element on in vitro plant tissue cultures (Winarto and da Silva 2015; Prando et al. 2014; Prades et al. 2012; Peixe et al. 2007). Its use during mushroom cultivation is not so popular, however on the studies of Zurbano et al. (2017), Kalaw et al. (2016), Jacob et al. (2015) and Magday et al. (2014) coconut water had been successfully used for cultivating *Pleurotus djamor, P. salmoneostramineus, Ganoderma lucidum* and *Lentinus* sp. Although the use of coconut water has not been fully contemplated for mushroom cultivation, it is considered as an important element during domestication of wild fungi according to Kalaw et al. (2016) and Magday et al. (2014). Above sated results coincide to the studies of Jacob et al. (2015), Prando et al. (2014) and Prades et al. (2012) that among the components of coconut water, sugars, minerals, free amino acids and growth promoting factors are considered those that makes it a suitable growth medium for microorganisms, plants and mushrooms.

In the same way, the present study also revealed that sweet potato decoction showed effect better than that of rice wash as medium for pure culture of *V. volvacea*. In this regard, the study of Nootjaree (2016) and Ubogu (2018), conforms and pointed out that the potential of yams as an efficient substrate for isolating mushroom from the fruiting bodies. Same analysis on the study of Amadi, et.al. (2012) showed that the purple sweet potato dextrose agar and purple sweet glucose agar can compete favorably with the common potato dextrose agar as medium for cultivation of fungi. Moreover, the alternative media of tubers decoction such as taro and cassava influences the growth of *Candida albicans* and *V. volvacea* compared to other alternative media as pointed out by Khusnul, et.al. (2020). Futher, on the study of Ubogu (2018) on the growth assessment of *Trichoderma viride, Aspergillus niger* and *Penicillium* sp on formulated culture medium, shows that the local tubers such as cocoyam, sweet potato, cassava and yam can be significantly used for fungal inoculation which was similar to the result of study of Wongjiratthiti and Yottakot (2017). Furthermore, the study of Zurbano, et.al (2017), revealed that the sweet potato sucrose gulaman recorded the widest mycelial run in the first four days compared with potato dextrose agar. This implies that sweet potato is efficient in developing pure culture of V. volvacea as supported by Nootjaree (2016), Ubogu (2018) and Amadi, et.al (2012).

Yield Performance of *V. volvacea* under Different Nutrient Sources and Substrates

The mean yields of *V. volvacea* with different nutrient sources and substrates in Siniloan, Laguna set-up are presented in Table 5. The mean yields without nutrient supplement were 659.54 grams, 669.15 grams with urea, and 611.03 with vermicast. For substrates, the lowest observed mean yield was 462.84 grams with wood shavings and the highest was 720.32 grams with banana leaves.

Analysis of variance did not show significant differences among mean yields without and with nutrient supplements in substrates. This means that the nutrient supplementation did not affect the yield of *V. volvacea.* On the other hand, significant differences were observed among the mean yields of mushroom grown in different substrates. Pairwise comparison shows that there were no significant differences among the mean yields with rice straw, corn bagasse, water hyacinth and banana leaves, however, with these four substrates, the mushroom yields were significantly higher than the yield with wood shavings. This result means that substrate materials are a significant factor that affect the yield of *V. volvacea*. No significant interaction between nutrient source and substrate was observed.

Table 5. Mean yields of *V. volvacea* with different nutrient sources and substrates in Siniloan, Laguna set-up, grams

Nutrient Source	Substrates					Mean
	Rice Straw	Wood Shavings	Corn Bagasse	Water Hyacinth	Banana Leaves	
No nutrient supplement	750.05	468.04	638.00	703.00	738.63	659.54[a]
Urea	699.26	466.10	713.63	699.71	767.03	669.15[a]
Vermicast	656.26	454.39	642.09	647.13	655.30	611.03[a]
Mean	701.86[a]	462.84[b]	664.57[a]	683.28[a]	720.32[a]	646.57

Analysis of variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	1700753.58	425188.40	30.77	0.00**
Nutrient	2	48518.38	24259.19	1.76	0.18ns
Substrate	4	658829.70	164707.43	11.92	0.00**
Nutrient x Substrate	8	35711.55	4463.94	0.32	0.95ns
Error	56	773726.28	13816.54		
Total	74	3217539.50			
CV (%)	18.18				

ns-not significant * - *significant* ** - *highly significant*

For Lumban, Laguna experimental set-up, the mean yields of *V. volvacea* are presented in Table 6. The mean yields were 636.64 grams, 666.16 grams and 678.72 with no nutrient supplement, with urea and with vermicast, respectively. With different substrates the mean yields ranged from 446.93 grams (wood shavings) to 734.71 grams (banana leaves).

No significant differences among nutrient supplement treatment means while significant differences were noted among substrate treatment means as shown by analysis of variance. Test for specific treatment differences showed that the yield with wood shavings was significantly lower than the mean yields with the other four substrates. These results mean that nutrient supplement is not a factor while substrate material is a factor of yield of Volvariella mushroom. Nutrient supplement-substrate interaction effect was not significant. It should be noted that these results for Lumban set-up were similar with the results for Siniloan set-up. The grand mean for Siniloan set-up was 646.57 grams which is lower than that of Lumban set-up with 660.51 grams

Table 6. Mean yields of *V. volvacea* with different nutrient sources and substrates in Lumban, Laguna set-up, grams

Nutrient Source	Substrates					Mean
	Rice Straw	Wood Shavings	Corn Bagasse	Water Hyacinth	Banana Leaves	
No nutrient supplement	701.07	432.95	672.65	684.51	692.03	636.64[a]
Urea	703.53	459.61	702.99	722.22	742.44	666.16[a]
Vermicast	735.35	448.22	722.62	717.72	769.67	678.72[a]
Mean	713.3[a]	446.93[b]	699.42[a]	708.15[a]	734.71[a]	660.51

Analysis of Variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	1915674.03	478918.51	58.83	0.00**
Nutrient	2	23324.70	11662.35	1.43	0.25ns
Substrate	4	865450.53	216362.63	26.58	0.00**
Nutrient x Substrate	8	8217.82	1027.23	0.13	0.99ns
Error	56	455867.75	8140.50		
Total	74	3268534.83			
CV (%)	13.66				

ns-not significant * - *significant* ** - *highly significant*

According to Kamaliah et al. (2022), the production of *V. volvacea* was shown to be highly impacted by bed temperature, humidity, replication, and harvesting week and month, but not by bed pH or orientation. A number of critical elements impacting mushroom production include temperature and humidity variations, especially during incubation, mycelial growth, fruiting body development, and the harvesting phase. According to the study, lowland dipterocarp forests offer the best conditions for growing edible mushrooms.

Growth Characteristics of *V. volvacea* under Different Nutrient Sources and Substrates

Number of Days from Mycelial Emergence to Pin head Formation

The mean number of days of mycelial emergence to pin head formation of *V. volvacea* grown under different sources of nutrients and substrate in Siniloan, Laguna is presented in Table 7. The longer period of mycelial emergence was observed on no nutrients were 15.40, urea were 15.80 days and 16.12 days with vermicast before the full colonization. For substrates, the shortest mycelial colonization was observed in banana leaves with 15.00 days and the longest period was 16.33 with water hyacinth.

Analysis of variance did not show significant differences among treatment means on number of days of mycelial emergence to pin head formation without and with nutrient supplements in substrates. This means that nutrient supplementation did not affect the mycelial emergence to pin hear formation of *V. volvacea.* Pairwise comparison shows that there were no significant differences among the mean number of days of mycelial emergence to pin head formation with rice straw, wood shavings, corn bagasse, water hyacinth and banana leaves. No significant interaction between nutrient sources and substrate was observed.

Table 7. Average number of days for mycelial emergence to pin head formation of *V. volvacea* under different nutrient sources and substrates in Siniloan, Laguna

Nutrient Source	Substrates					Mean
	Rice straw	Wood shavings	Corn Bagasse	Water hyacinth	Banana leaves	
No nutrient supplementation	15.00	15.00	15.80	15.80	15.40	15.40
Urea	15.80	16.40	15.40	16.20	15.20	15.80
Vermicast	16.00	16.80	16.40	17.00	14.40	16.12
Mean	15.60	16.07	15.87	16.33	15.00	

Analysis of variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	11.5467	2.8867	1.77	0.15ns
Nutrient	2	6.5067	3.2533	2.00	0.14ns
Substrate	4	15.5467	3.8867	2.39	0.06ns
Nutrient x Substrate	8	14.2933	1.7867	1.10	0.38ns
Error	56	91.2533	1.6295		
Total	74	139.1467			

ns-not significant * - *significant* ** - *highly significant*

For Lumban set-up, data are presented in Table 8. The longer period of mycelial emergence was observed on no nutrients which is 13.12, urea with 13.12 and 13.00 with vermicast before the full colonization. For substrates, the shortest mycelial colonization was observed in rice straw, wood shavings, and banana leaves with 13.00 days and the longest period was 13.33 with corn bagasse.

Analysis of variance did not show significant difference among treatment mean numbers of days of mycelial emergence to pin head formation without and with nutrient supplements in substrates. This means that nutrient supplementation did not affect the mycelial emergence to pin hear formation of *V. volvacea.* Pairwise comparison shows that there were no significant differences among the mean numbers of days of mycelial emergence to pin head formation with rice straw, wood shavings, corn bagasse, water hyacinth and banana leaves. No significant interaction between nutrient sources and substrate was observed.

Table 8. Average number of days for mycelial emergence to pin head formation of *V. volvacea* under different nutrient sources and substrates in Lumban, Laguna

Nutrient Source	Substrates					Mean
	Rice straw	Wood shavings	Corn Bagasse	Water hyacinth	Banana leaves	
No nutrient supplementation	12.80	13.20	13.80	13.40	12.40	13.12
Urea	12.00	12.80	13.80	13.80	13.20	13.12
Vermicast	14.20	13.00	12.40	12.00	13.40	13.00
Mean	13.00	13.00	13.33	13.07	13.00	

Analysis of variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	9.5200	2.3800	0.99	0.42^{ns}
Nutrient	2	0.0000	0.0000	0.00	1.00^{ns}
Substrate	4	1.6533	0.4133	0.17	0.95^{ns}
Nutrient x Substrate	8	25.8667	3.2333	1.34	0.24^{ns}
Error	56	134.8800	2.4086		
Total	74	171.9200			

ns-not significant * - *significant* ** - *highly significant*

This implies that experimental units in Lumban, Laguna had shorter period of mycelial inoculation compared to those of Siniloan, Laguna. On the study of Thuc (2020), it was pointed that the emergence of mycelia after incubation period was 14-15 days and the pin head formation shall constitute 5 days after emergence. The optimal temperature favorable for mycelial growth was 30-35℃ and 20-30℃ for its growth and development.

In the present study, the mycelial growth to pinhead formation was affected by the temperature requirements and relative humidity. Reflecting on Table 2 (climatic requirement), the Tmax of both location coincides on the optimal temperature requirements for mycelial growth showing mean Tmax of 32.92℃ for Siniloan and 30.41℃ in Lumban, Laguna respectively. However, the longer the period of mycelial emergence to pin head formation was affected by the relative humidity which the experimental area located in Siniloan, Laguna has 79.25% and 79.92% in Lumban, Laguna, respectively. This claims contradicts on the study of Marroquin (2017), that mycelial growth increased significantly with increasing RH from 81.5% RH.

Pin head size of V. volvacea with different nutrient sources and substrates

The mean pin head size of *V. volvacea* with different nutrient sources and substrates in Siniloan, Laguna set-up are presented in Table 9. The mean pin head size without nutrient were 27.33 mm, 27.78mm with urea and 27.49mm with vermicast. For substrate, the highest observed pin head size was 28.01mm with corn baggase and lowest was 26.92mm with wood shavings.

Analysis of variance did not show significant difference among mean pinhead size without and with nutrient supplements in substrates. This means that nutrient supplementation did not affect the pin head size of *V. volvacea.* Pairwise comparison shows that there were no significant differences among the mean pin head size with rice straw, wood shavings, corn baggase, water hyacinth and banana leaves. No significant interaction between nutrient sources and substrate was observed.

Table 9. Mean pinhead size (mm) of *V.volvacea* in Siniloan, Laguna grown under substrates supplemented with different nutrient sources.

Nutrient Source	Substrates					Mean
	Rice straw	Wood shavings	Corn Bagasse	Water hyacinth	Banana leaves	
No nutrient supplementation	25.86	26.54	28.88	27.33	28.04	27.33
Urea	28.14	25.92	28.48	28.95	27.40	27.78
Vermicast	27.43	28.31	26.68	27.38	27.64	27.49
Mean	27.14	26.92	28.01	27.89	27.69	

Analysis of variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	7.3752	1.8438	0.36	0.83^{ns}
Nutrient	2	5.2335	2.6168	0.52	0.60^{ns}
Substrate	4	5.5942	1.3986	0.28	0.89^{ns}
Nutrient x Substrate	8	46.4947	5.8118	1.15	0.34^{ns}
Error	56	282.8915	5.0516		
Total	74	347.5893			
CV (%)	8.12				

ns-not significant * - *significant* ** - *highly significant*

The mean pin head size of *V. volvacea* with different sources and substrates in Lumban, Laguna set-up are presented in Table 10. The mean pin head sizes were 28.90mm without nutrient, 28.13mm with urea and 27.06mm with vermicast. For substrates, the highest observed pin head size was 28.70mm with corn bagasse and lowest was 26.97mm with wood shavings.

Analysis of variance showed significant difference among mean pin head sizes without and with nutrient supplements in substrates. This means that nutrient supplementation affects the pin head size of *V. volvacea*. Likewise, significant differences were observed among mean pin head sizes of mushrooms grown in different substrates. A significant interaction effect between nutrient sources and substrates was observed. The highest pin head size was observed were 30.28mm with no nutrient and water hyacinth. The lowest pin head size was observed among mushrooms were 25.43mm supplemented with vermicast with water hyacinth.

Table 10. Mean pin head size (mm) of *V.volvacea* in Lumban, Laguna grown under substrates supplemented with different nutrient sources

Nutrient Source	Substrate					Mean
	Rice straw	Wood shavings	Corn Bagasse	Water hyacinth	Banana leaves	
No nutrient supplementation	27.23^{a}	27.52^{a}	29.80^{a}	30.28^{a}	29.68^{a}	28.90
Urea	26.83^{a}	26.38^{a}	29.12^{ab}	29.59^{a}	28.71^{ab}	28.13
Vermicast	28.36^{a}	27.01^{a}	27.19^{b}	25.43^{b}	27.30^{b}	27.06
Mean	27.47	26.97	28.70	28.43	28.56	

Mean with the same letter are not significantly different.

Analysis of variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	15.4855	3.8714	1.11	0.36^{ns}
Nutrient	2	42.8109	21.4055	6.13	0.00**
Substrate	4	35.0161	8.7540	2.51	0.05*

Nutrient x Substrate	8	68.2839	8.5355	2.44	0.024*
Error	56	195.6217	3.4932		
Total	74	357.2182			
CV (%)	6.67				

ns-not significant * - *significant* ** - *highly significant*

Analysis revealed that significant effects of nutrient supplement and substrate interaction were observed in in Lumban Laguna set-up. However, irrespective of substrates and nutrient supplementation used pin head sizes were significantly the same. This finding conforms to the studies of Ahlawat (2003), Krishnamoorthy (2005), Royse (2014), Bao (2010) and Poppe (2000). Rice straw can be compared to wood shavings, dried banana leaves, corn bagasse, and water hyacinth. Like corn bagasse, rice straw, water hyacinth, and dried banana leaves, wood shavings also include cellulose, hemicellulose, and lignin, which are crucial for the development of straw mushrooms. These substrates give the mushroom's mycelium a good carbon source from which to grow and develop. Dried banana leaves can disintegrate gradually over time, as can maize bagasse, rice straw, and water hyacinth. The substrate is gradually decomposed, releasing nutrients that the mushroom mycelium can use to develop and fruit. The decomposition contributes to the creation of an ideal habitat for the life cycle of the mushroom. Additionally, these materials have adequate porosity to permit optimal aeration and moisture retention, both of which are essential for efficient mushroom cultivation. This implies that any substrates that contains essential properties is suitable to *V. volvacea* production according to the similar studies conducted to other mushroom cultivars by Ahlawat and Kumar (2005), Onuaha (2008) and Bao et.al. (2013).

Length of Stipe with different nutrient sources and substrates

For Siniloan set-up, the mean stipe lengths of *V. volvacea* with different nutrient sources and substrates are presented in Table 11. The mean stipe length without nutrient supplement were 7.71cm, 8.39cm with urea and 8.68cm with vermicast. For substrates, the lowest mean stipe length observed was 6.71cm with wood shavings and the highest was 8.89cm with banana leaves.

Analysis of variance shows significant differences among mean stipe lengths without and with nutrient supplements in substrates. This means that the nutrient supplementation affects the stipe length of *V. volvacea.* Also, significant differences were observed among the mean stipe lengths of mushroom grown in different substrates. Pairwise comparison shows that there were no significant differences among the mean stipe length with rice straw, corn bagasse, water hyacinth and banana leaves, however the mean stipe lengths with these four substrates were significantly higher than that of wood shavings. Moreover, there were significant difference among nutrient sources. This result means that substrate materials are a significant factor that affect the stipe length of *V. volvacea.* No significant interaction between nutrient source and substrate was observed.

Table 11. Average stipe length (cm) of *V.volvacea* in Siniloan, Laguna grown under substrates supplemented with different nutrient sources.

Nutrient Source	Substrates					Mean
	Rice straw	Wood shavings	Corn Bagasse	Water hyacinth	Banana leaves	
No nutrient supplementation	7.72	7.34	7.35	7.96	8.16	7.71[b]
Urea	8.73	6.49	8.96	8.64	9.15	8.39[ab]
Vermicast	8.87	6.31	9.26	9.60	9.35	8.68[a]
Mean	8.44[a]	6.71[b]	8.52[a]	8.73[a]	8.89[a]	

Mean with the same letter are not significantly different.

Analysis of variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	6.5176	1.6294	0.88	0.48[ns]
Nutrient	2	12.5516	6.2756	3.38	0.04*
Substrate	4	46.71.82	11.6795	6.28	0.00**
Nutrient x Substrate	8	15.8570	15.8570	1.07	0.40[ns]
Error	56	104.0669	1.8583		
Total	74	185.7109			
CV (%)	16.50				

ns-not significant * - *significant* ** - *highly significant*

The mean stipe lengths for Lumban set-up of *V. volvacea* with different nutrient sources and substrates are presented in Table 12. The mean stipe length without nutrient supplement were 7.99cm, 8.30cm with urea and 8.30cm with vermicast. For substrates, the lowest mean stipe length observed was 6.23cm with wood shavings and the highest was 9.20cm with banana leaves.

Analysis of variance did not show significant differences among mean stipe length without and with nutrient supplements in substrates. This means that the nutrient supplementation did not affect the stipe length of *V. volvacea.* On the other hand, significant differences were observed among the mean stipe length of mushroom grown in different substrates. Pairwise comparison shows that there were no significant differences among the mean stipe length with water hyacinth and banana leaves and between rice straw and corn bagasse. However, the first pair had significantly higher mean than the second pair. The stipe length in wood shavings was lowest. These results means that substrate materials are a significant factor that affect the stipe length of *V. volvacea*. No significant interaction between nutrient source and substrate was observed.

Table 12. Mean stipe length (cm) of *V.volvacea* in Lumban, Laguna grown under substrates supplemented with different nutrient sources.

Nutrient Source	Substrates					Mean
	Rice straw	Wood shavings	Corn Bagasse	Water hyacinth	Banana leaves	
No nutrient supplementation	7.94	6.48	8.11	8.70	8.74	7.99[a]
Urea	8.22	6.41	8.12	9.17	9.56	8.30[a]
Vermicast	8.80	5.80	7.94	9.70	9.28	8.30[a]
Mean	8.32[b]	6.23[c]	8.06[b]	9.19[a]	9.20[a]	

Means with the same letter are not significantly different.

Analysis of variance

Source	DF	Sum of Square	Mean of Square	F value	P-value
Block	4	11.0263	2.7566	3.31	0.02*
Nutrient	2	1.5448	0.7724	0.93	0.40ns
Substrate	4	88.4373	22.1093	26.51	0.00**
Nutrient x Substrate	8	6.1144	6.1144	0.7643	0.51ns
Error	56	46.7001	0.8339		
Total	74	153.8229			
CV (%)	11.14				

ns-not significant ** - significant* *** - highly significant*

The longest stipes were produced in banana leaves and water hyacinth followed by rice straw and corn bagasse, the shortest were those from wood shavings substrate. Evidently, on the study of Hyunjong and Byung (2004) supported by Teddese (2012) they pointed out that banana leaves is comparable to rice straw in growing *V. volvacea* with high yield in the entire cycle. No significant interaction effects of variables were observed. Also, Chaisaena, et.al (2012) pointed out that an existing variety of lignocellulose agricultural wastes after harvesting and supply plant weeds can be used for mushroom production. It was determined on their study that the suitability of solid substrates such as water hyacinths, rice straw and sunflower residues for growing these edible mushrooms is efficient. This means that nutrient sources and substrates influenced the length of stipes independently.

Proximate Analysis using Kjeldahl Distillation Method in Determining the Total Nitrogen Content and Protein (%) in *V. volvacea* with Different Nutrient Sources and Substrates

Nitrogen contents resulting from Kjeldahl distillation method and computed protein contents of *V. volvacea* are shown in Table 13. Analysis revealed that the highest N content was found on Wood shavings supplemented with vermicast with 4.34%N and the lowest N content was found in water hyacinth without nutrient supplementation with 3.07%N.

On the other hand, data on protein content was analyzed by converting the Total N multiplied by 6.25. Conversion of nitrogen shows that the higher protein content was found in Wood shavings substrate supplemented with vermicast that contains 27.13%. Meanwhile, the lowest protein content was found in *V. volvacea* grown in water hyacinth without supplementation containing 19.19% protein.

The conversion of %N to %Protein was undertaken because mushroom is claimed as substitute to meat and maybe fish as source of protein. The assertion that mushrooms, regardless of their size or form, can offer the same amounts of nutrients per serving is supported by Ware (2019). It also showed that protein content of mushroom is 3.09g in 100g cup of mushrooms which can be taken 46-56g daily the same as meat with 21.9-23.1g of protein intake daily. In addition, it indicates the daily allowance of each nutrient for individuals based on age and sex.

Table 13. Analysis of Total Nitrogen Content of dried *V. volvacea* under different nutrient sources and substrates.

Nutrient Sources and Substrate Combination	Nitrogen Content, % (w/w)	Protein Content (%)
No nutrient supplement, Rice Straw	3.96 ± 0.05	24.75
No nutrient supplement, Wood shavings	4.13 ± 0.01	25.81
No nutrient supplement, Corn bagasse	4.32 ± 0.01	27.00
No nutrient supplement, Water hyancinth	3.07 ± 0.10	19.19
No nutrient supplement, Banana leaves	2.72 ± 0.01	17.00
Urea, Rice Straw	3.49 ± 0.02	21.81
Urea, Wood shavings	3.60 ± 0.01	22.50
Urea, Corn bagasse	4.23 ± 0.07	26.44
Urea, Water hyancinth	3.99 ± 0.06	24.94
Urea, Banana leaves	3.39 ± 0.10	21.19
Vermicast, Rice Straw	3.92 ± 0.05	24.50
Vermicast, Wood shavings	4.34 ± 0.01	27.13
Vermicast, Corn bagasse	4.28 ± 0.06	26.75
Vermicast, Water hyancinth	4.00 ± 0.01	25.00
Vermicast, Banana leaves	3.96 ± 0.03	24.75

The nitrogen contents were expressed as a dried weight using Sangthong et al. (2022) which showed 8.15 ± 0.28 as the moisture content. From *V. volvacea*, the following content was extracted: 43.16 ± 0.38% for carbohydrates, 19.40 ± 0.29% for protein, 2.49 ± 0.18 for crude fat, 11.71 ± 0.28 for ash, and 15.10 ± 0.16 for fiber. On the study of Haq et.al (2011), it pointed out that the biochemical analysis of the fruiting bodies obtained showed that fruiting bodies harvested from different substrates varied significantly in nitrogen, crude protein, ash, and moisture percentage. It revealed that nitrogen percentage of the fruiting bodies harvested from cotton waste was the highest (5.56%) followed by the paddy straw (4.87%) and sugarcane baggasse (4.45%). The fruiting bodies harvested from corn stovers and banana leaves had almost same nitrogen percentage. The lowest percentage of nitrogen was found in pulses straw (3.22%).The protein contents were maximum in the fruiting bodies harvested from cotton waste (34.17%) followed by the sugarcane baggasse (30.51%), paddy straw (28.57%), banana leaves (23.92%), corn stovers (21.77%) and pulses straw (20.25%).

Biological Efficiency of Substrates Supplemented with Different Nutrient Sources

The mean biological efficiency of substrates supplemented with different nutrient sources are presented in Table 14. The mean percentage without or with nutrient supplement were 25.40%, 25.28% with urea and 21.34% with vermicast. For substrates, the highest biological efficiency observed was 29.37% with banana leaves and the lowest was 13.11% with wood shavings.

Analysis of variance shows significant differences among mean percentages without or with nutrient supplements in substrates. This means that the nutrient supplementation affects the biological efficiency of substrates. In the same way, significant differences were observed among the mean biological efficiencies with different substrates. Pairwise comparison shows that there were no significant differences among the mean biological efficiencies with rice straw, water hyacinth and banana leaves and the biological efficiencies with these three substrates were significantly higher than those with corn bagasse and wood shavings. Biological efficiency was lowest with wood shavings. No significant interaction between the nutrient sources and substrates was observed.

Table 14. Biological efficiency (%) of substrates supplemented with different nutrient sources.

Nutrient Source	Substrates					Mean
	Rice straw	Wood shavings	Corn Bagasse	Water hyacinth	Banana leaves	
No nutrient supplementation	28.10	13.22	23.98	29.96	31.73	25.40[a]
Urea	30.88	12.69	22.17	28.41	32.23	25.28[a]
Vermicast	27.62	13.41	19.08	22.47	24.14	21.34[b]
Mean	28.87[a]	13.11[c]	21.74[b]	26.95[a]	29.37[a]	

Analysis of variance

Source	DF	Sum of Square	Mean Square	F value	P-value
Replication	4	3011.80	752.95	20.79	0.00**
Nutrient Source	2	265.78	132.89	3.67	0.03*
Substrate	4	2773.81	693.45	19.15	0.00**
Nutrient Source x Substrate	8	189.90	23.74	0.66	0.73ns
Error	56	2028.28	36.22		
Total	74	8269.57			

ns-not significant ** - significant* *** - highly significant*

V. volvacea depends largely on the cellulose and hemicellulose of the substrates, their total nitrogen and carbon contents and C:N ratio. Presence of more cellulose content and higher C:N ratio in banana leaves, rice straw and water hyacinth substrates could be one of the reason for higher yield and biological efficiency of *V.volvacea*. This finding further confirming the reports of Biswas and Layak (2014). Compactness and high lignin to substrates may lead to the poor formation of mushroom and hence reduced the yield. Similar observations with wheat straw substrate made by Gracha and Kalra (1978) which was confirmed by Biswas and Layak (2014). Further, the result of the present study contradicts on the established biological efficiency. On the work of Belewu and Belewu (2005) and Zikriyani et.al (2018) it showed that the range of biological efficiency of banana leaves substrate is 8.6%-15.20% while in the present study it revealed 29.37%. Moreover, in the work of Chang et.al (1982), Miles (2008), and Biswas (2014) they identified that the biological efficiency of rice straw to provide high production is ranging from 10.2%-15.00% compare to 28.87%. Moreso, water hyacinth obtained 8.7% as Thiribhuvanamala et al. (2012) stated on their study on improving the biological efficiency of *V. volvacea* which lower to the present study obtained 26.95% biological efficiency. This means that the lower the biological efficiency the higher the production of *V. volvacea*.

Economic potential of *V. volvacea* Production with Different Nutrient Sources and Substrates

To facilitate in decision-making which particular technology the farmer can use, the cost and return is presented in Table 15. The data presented were based on the actual production in this study. The evaluation of costs and returns done by substrate materials and nutrient sources.

Among the substrates used in the study, the return on invest of banana leaves (27.51%), rice straw (22.22%), water hyacinth (21.31%) and corn baggase (13.74%) are comparable even without nutrient supplementation, respectively. However, in the case of Wood shavings substrate, though it produced *V. volvacea*, it is deemed not suitable for the production with return on investment of -27.67%.

On the other hand, the highest return on investment under substrates supplemented with inorganic fertilizer (urea) was found in the banana leaves (25.42%) while the lowest was the wood shavings substrate with ROI of -31.37%. Moreover, potential of corn bagasse substrate supplemented with vermicast showed the highest return on investment of about 9.23% an the lowest was the Wood shavings substrate with return on invest of -33.62%. Banana leaves gave the highest ROI except when vermicast was used as nutrient source.

This implies that any of the four (4) substrates can be used with or without nutrient supplementation as indicated on Table 5 of this study. However, for economic purposes the farmer may use these results as basis in deciding which among the substrates they will use. Further, the wood shavings substrate, was proven for *P. ostreatus* production as efficient substrate materials and widely used in the local community.

Table 15. Comparative economic potential of *V. volvacea* under different nutrient sources and substrates.

Nutrient Source	Indicators	Substrates				
		Rice straw	Wood shavings	Corn baggase	Water Hyacinth	Banana leaves
No Nutrient Source	Sales	1,125.00	702.00	1,047.00	1,056.00	1,110.00
	Expenses	920.50	970.50	920.50	870.50	870.50
	Net Income	204.50	-268.50	126.50	185.50	239.50
	ROI (%)	22.22	-27.67	13.74	21.31	27.51
Urea	Sales	1,050.00	699.00	1,071.00	1,050.00	1,152.00
	Expenses	968.50	1,018.50	968.50	918.50	918.50
	Net Income	81.50	-319.50	102.50	131.50	233.50
	ROI (%)	8.42	-31.37	10.58	14.32	25.42
Vermicast	Sales	984.00	684.00	1,881.00	972.00	984.00
	Expenses	980.50	1,030.50	980.50	930.50	930.50
	Net Income	3.50	-346.50	90.50	41.50	53.50
	ROI (%)	0.36	-33.62	9.23	4.46	5.75

CONCLUSIONS

Based from the data presented, it is concluded that sweet potato decoction is efficient growing media for pure culture of *V. volvacea* inoculation. Also, grown *V. volvacea* produces same yield regardless of the nutrient sources applied to locally available substrates such as banana leaves, rice straw, corn bagasse and water hyacinth. Its mycelial emergence to pin head formation is not dependent on the nutrient source and substrate material. Pin head size may or may not be affected by nutrient supplementation and kind of substrate depending on the conditions in the culture areas or locations. The length of stipe is affected by nutrient source and kind of substrate. Mushroom tissues with dried tissues gave higher N content and protein content when organic nutrient source is used. Substrates that are easily decomposed gave higher biological efficiency. Return on investment determined by nutrient supplementation and substrate materials. The effect is primarily based on the ability of the substrate to support higher yield as the materials are locally available.

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