COMPARISON ON GROWTH AND YIELD OF WHITE OYSTER MUSHROOM (*Pleurotus florida*) ON WASTE PAPER PULP AND SAWDUST AS SUBSTRATES

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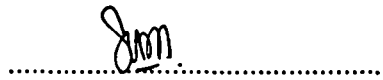

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COMPARISON ON GROWTH AND YIELD OF WHITE OYSTER MUSHROOM (Pleurotus florida) ON WASTE PAPERR PULP AND SAWDUST AS SUBSTRATES

ABSTRACT

Pleurotus florida or commonly known as White Oyster Mushrooms are widely and commercially grown for food source. In this experiment, *P.florida* will be grown on waste papers pulp as the substrate with the addition of gypsum, rice bran and rice husk as the supplement in a fixed ratio. Sawdust substrate will be used as the control in this study and there are 6 replications in each treatments while there were 6 treatment in this experiment which will the bring total of 30 replications. The growth and development of fruit body were observed. Parameters that were tested in this experiment are mycelium run rate, numbers of day for primordia and fruiting bodies formation, fresh weight of fruit bodies and biological efficiency. Based on the result obtained, mycelium run rate showed no significant difference ($F_{5,\ 35}$=2.348, P>0.05) in all treatments while formation of primordia and fruiting bodies showed significat difference ($F_{5,\ 35}$=66.874, P<0.05) and ($F_{5,\ 35}$= 9.258, P<0.05). Diameter and fresh weight of fruiting bodies also result in significant difference ($F_{5,\ 35}$= 10.517, P < 0.05) and ($F_{5,\ 35}$= 10.692, P<0.05). There was no huge difference in biological efficiency for all substrate bags.

PERBANDINGAN PERTUMBUHAN DAN HASIL CENDAWAN TIRAM PUTIH MENGGUNAKAN PULPA KERTAS TERPAKAI DAN HABUK KAYU SEBAGAI SUBSTRAT

ABSTRAK

Pleurotus florida atau dikenali sebagai Cendawan Tiram Putih digunakan secara meluas dan ditanam untuk tujuan komersial sebagai sumber makanan. Dalam eksperimen ini, pulpa kertas terpakai akan digunakan bagi tujuan penanaman P.florida sebagai substrat dengan menambah kapur, dedak padi dan kulit padi sebagai supplemen dalam nisbah yang ditetapkan. Substrat habuk kayu digunakan sebagai kawalan dan terdapat 6 replikasi dalam setiap rawatan.Terdapat 6 rawatan dalam eksperimen ini. Oleh itu, jumlah kesemua replikasi adalah 30. Pemerhatian terhadap pertumbuhan dan perkembangan jasad buah dibuat. Parameter yang telah diuji dalam kajian ini adalah kadar pertumbuhan miselium, pembentukkan primordia dan jasad buah dalam bilangan hari, berat basah dan diameter jasad buah dan juga keberkesanan biologikal. Berdasarkan keputusan yang diperoleh, kadar pertumbuhan miselium tidak menghasilkan sebarang perbezaan ketara ($F_{5,\ 35}$=2.348, P>0.05) dalam setiap rawatan manakala pembentukkan primordia dan jasad buah menunjukkan perbezaan ketara iaitu ($F_{5,\ 35}$=66.874, P<0.05) and ($F_{5,\ 35}$= 9.258, P<0.05). Diameter dan berat basah cendawan turut menunjukkan perbezaan ketara iaitu ($F_{5,\ 35}$= 10.517, P < 0.05) and ($F_{5,\ 35}$= 10.692, P<0.05). Tiada perbezaan yang dicatatkan bagi keberkesanan biologikal dalam setiap beg substrat.

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LIST OF SYMBOLS UNITS AND ABBREVIATION

%	Percentage
spp	Species
EFB	Empty Fruit Bunch
^0c	Degree Celsius
PDA	Potato Dextrose Agar
MC	Moisture Content
PVC	Poly Vinyl Chloride
g	Gram
cm	Centimetre
C/N	Carbon/Nitrogen

LIST OF FORMULA

Formula	Page
$\text{Biological efficiency} = \frac{\text{Fresh weight of fruit bodies}}{\text{Dry weight of substrate}} \times 100$	16

CHAPTER 1

INTRODUCTION

1.1 Background

Mushroom are fleshy, spore-bearing reproductive structures of fungi which is grown on organic substrate and had played an important role in human dietary due to its nutritional benefits (Etich, 2013). The growth and production of mushrooms are different from green plants which fully depend on sunlight to produce its own food through photosynthesis process while mushrooms rely on organic material which also act as substrate for their nutrition. This is because they are lack of chlorophyll and therefore cannot produce their own food. This characteristic of mushroom are known as absorptive heterotroph organisms which enable them to grow on decompose organic materials.

There are more than 2000 type of wild mushroom and only less than 25 mushrooms are considered as an edible mushroom. Example of wild edible mushrooms are *Agaricus bitorquis*, *Pleurotus ostreatus*, *Volvariella volvaceae* and *Lentinula edodes*. These type of mushrooms had been cultivated widely for commercial purposes (Barros *et al.*, 2007). General morphology of mushrooms consist of stalk, cap, fruit-body and gill which is located under the cap of the fruit-body of mushroom.

Mushrooms are a good source of protein, vitamins and mineral and widely known to have a broad range of uses in food culinary and medicinal purposes. Nutrition values of Oyster mushroom had been reported contain with protein (25-50%), sugars (17-47%), mycocellulose (7-38%) and minerals such as potassium, phosphorus, calcium and sodium about 8-12% (Stanley, 2011). Edible mushrooms also rich in vitamins like niacin, riboflavin, vitamin D, C, B1, B5 and B6 (Syed and Kadam, 2009).

Mushroom species from the genus of *Pleurotus* is very popular in South-east Asian countries where it can be grown all around the year. *Pleurotus* species are characterized by the rapidity of the mycelia growth and high saprophytic colonization activity on cellulosic substrate. They have the ability to breakdown cellulose and lignin bearing materials without composting (Kapoor, 2010). There are many species under the genus of *Pleurotus* such as *Pleurotus ostreatus*, *Pleurotus erygii* and *Pleurotus sajor caju*.

The White Oyster Mushroom or its scientific name *Pleurotus florida* so far is the best flavor of the oyster mushrooms. The thick and firm stems have a pleasant texture and a slightly sweet taste. Another plus point is, its long storage life compared to other oyster mushroom varieties. This variety can easily be transported over long distances. It has also recently become known that this variety, just like the shiitake, lowers cholesterol levels and an excellent selling point in cultures where cardio-vascular diseases are major killers.

Most of *Pleurotus* spp can be grown on various types of substrates including paddy straw, maize stalk, vegetable plant residues, baggase etc. It also can be cultivated by using industrial wastes like paper mill/pulp residues, coffee by-products, and tobacco waste. The cultivation of *Pleurotus* spp in Malaysia mostly using agricultural wastes such as Empty fruit bunches (EFB) of oil palm, paddy straw, sawdust and banana leaves as substrates. Agricultural wastes which contain high contain of lignin and cellulose are potential to be alternatives substrate for mushroom cultivation.

1.2 Justification of study

Cultivation of mushroom can play an important role in managing not only agricultural waste but also industrial waste whose disposal had become major threat to the worldwide environment. Recycling industrial waste like paper pulp into usefull and environmental friendly materials. In Malaysia, it also can act as alternative substrate replacing agricultural wastes such as oil palm's empty fruit bunches (EFB) and paddy straw in cultivating mushroom in the future. Paper waste pulp contain high carbon and with the addition of nitrogen which can be utilized by most oyster mushroom.

1.3 Objective

The objective of this experiment was to evaluate the growth and yield of *Pleurotus florida* using waste paper pulp and sawdust as substrates.

1.4 Hypothesis

Ho: There was no significant difference in growth and yield of *Pleurotus florida* on paper waste pulp and sawdust as substrates.

Ha: There was no significant difference in growth and yield of *Pleurotus florida* on paper waste pulp and sawdust as substrates.

CHAPTER 2

LITERATURE REVIEW

2.1 Macro fungi

Fungi are the distinct group of organisms which include the species with large and visible fruiting bodies (macro-fungi). Mushrooms are the best example of macro-fungi. Mushrooms macro-fungi usually classified into *Basidiomycetes* and *Ascomycetes*. Mushrooms differ from any other plant as they cannot produce their own food (Chang and miles, 2004). Mushroom producing fungi serve valuable roles in ecosystem. They decompose and responsible for the recycling dead organic matter into molecular form which will be used by other organisms as source of nutrients.

Generally, they have a cap (pileus) and stalk and frequently seen to be grown in the field and forests. Most of the mushrooms growing on the forest floor are intimately linked to trees by symbiosis. This association, called mycorrhiza, occurs between the root ends of a tree and the vegetative system of a mushroom. Mycorrhiza benefits both organisms in which there is an exchange of nutrients. In general, the mushroom helps the tree extract minerals and water from the soil while the tree supplies the mushroom with sugar compounds (carbohydrates). Another mode of nutrition of fungi is saprophytism and parasitism.

Saprophytism is another important living method for mushrooms, especially for species which grow on lawns, on rotting wood or on excrement. It feeds itself by digesting the organic matter and at the same time returns nutrients to the soil. Some mushrooms are parasites. There are several kinds of parasitism which is the species that attacks a healthy host (tree, plant or insect) and lives on it without killing it or attacks only unhealthy hosts, thereby hastening their death. The parasitic species are generally microscopic mushrooms.

A mushroom has two parts. The underground part is called the mycelium. It gets food for the mushroom. The umbrella-shaped body of a mushroom is called the fruit or sporophore. The fruit starts out as a small button which grows into a stalk and a cap. The stalk or stem grows quickly because it can absorb a lot of water. Mushrooms can have a diameter of up to 40 cm. Most edible mushrooms are white, brown or yellow in colour. Typical mushroom and its general component are shown in the figure 2.1.

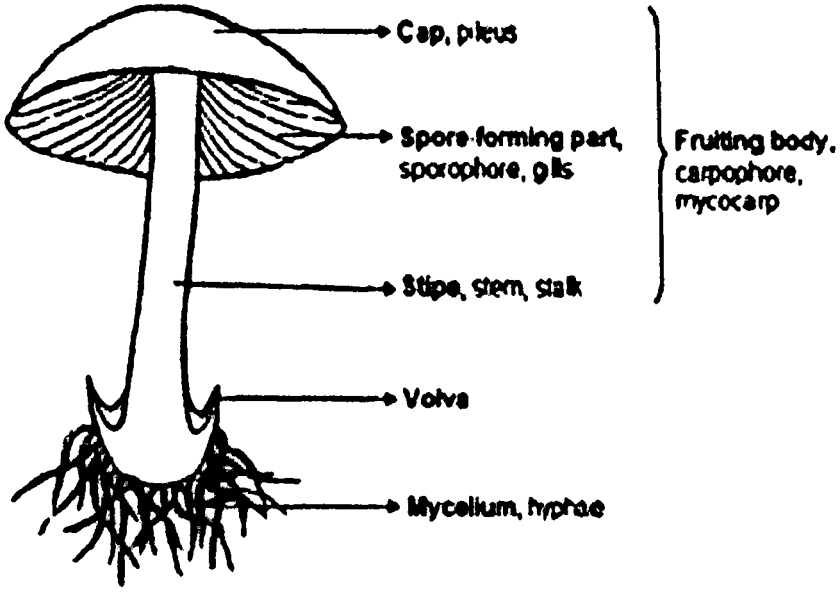

Figure 2.1 General components of a typical mushroom

Source: (Ren, Perera and Hemar, 2012)

The most active part of the mushroom component is the mycelium which live inside the substrate (wood, straw, grain, etc). Mushrooms absorb nutrient from its environment through collectively mass of hypae or mycelium in two stages process. The first process involve the secretion of enzyme from the hypae into decaying wood or substrate. Secondly, the enzyme will break down biological polymers into smaller units such as

monomers. The mycelium will absorb these monomers using a combination of facilitated diffusion and active transport. In this study, waste paper pulp substrate will be used as substrate with the addition of supplement to support the growth of mycelium.

2.2 Nutritional value and market demand of mushrooms

Mushroom has been known as a source of food since thousand years ago (Mattila *et al.*, 2001). Mushrooms had been widely used as food due to its delicacy and its nutritional value. They contain a large arrays of nutrients and any other natural phytochemicals that have wide ranges of nutritional and health benefits. Their benefits include wound-healing, immunity enhancement and tumor retarding effects (Cheung, 2010). Malnutrition gap in global population can be reduced due to its high protein and nutritive value of mushrooms (Dai and Yang, 2009). Their values also recently had been promoted to tremendous level with medicinal mushrooms trial conducted for HIV/AIDS patients in Africa which showed encouraging result (Chang, 2006).

In general, the fruiting bodies of mushrooms contain about 56.8% carbohydrate, 25% protein, 5.7% of fat and 12.5% ash on its dry basis (Ouzouni *et al.*, 2009). Edible mushrooms also contain several type phenolic compounds recognized as an excellent antioxidant due to its ability to scavenging free radicals released in the environment (Ribeiro *et al.*, 2006). The table 2.2 showed nutrition content of mushroom with different type of foods.

Source: Parjimo and Andoko, 2012.

Type of food	Protein (%)	Fat (%)	Carbohydrates (%)
Paddy straw mushroom	1.8	0.3	4
Oyster mushroom	27	1.6	58
Meat	21	5.5	0.5
Spinach	-	2.2	1.7
Potatoes	2	-	20.9
Cabbage	1.5	0.1	4.2
Peas	-	2.4	0.2

Table 2.2.1 Nutrition content of mushrooms with different type of food

The market demand of mushrooms have been increasing worldwide. In regional term, Asia and Europe accounted for approximately 99% of the market with North America contributing less than 0.1% to the overall total. In 2002, mushrooms products (mushrooms derivative from medicinal, edible and wild mushrooms) used mainly for dietary supplement (mushrooms nutriceuticals) was assessed to have generated about US$11 billion. The US market mushrooms for dietary supplements had been estimated approximately US$35 million in 1994.

The industry of mushrooms cultivation in Malaysia is known for source of food and health benefits. However, the cultivation of mushrooms in Malaysia is still new and small compared to any other agriculture crops industry. This industry however become one of the highest valued crops in Malaysia. The Malaysian government has categorized mushrooms as one of the seven high value crops in Malaysia. The land area for mushrooms cultivation had been increase with the government support. The demand for mushrooms in local as well as in the world had increase with time. This is reflected by the increase of importation of fresh mushrooms from 80.9 million tons in 2007 to more than 90.8 million tons in 2008 (FAO, 2009). The world demand for fresh mushrooms also is growing about 15% a year due to increase in population and consumption. In the local market, the daily demand for fresh mushrooms is around 50,000 kg while the supply is only 24,000 kg (MOA, 2010). The total demand for mushrooms is projected to increase from 23,000 tons per year to 72,000 tons per year in 2020.

However, mushrooms industry in Malaysia is still lacking of information on supply and demand of mushrooms product, consumers perception and preference towards fresh mushrooms. There is no much data are available for industry players and policy makers to make strategic decision on the production on marketing of fresh mushrooms in Malaysia. In 2008, the number of farmers and companies involve in the cultivation of mushrooms was more than 850. However, the numbers had been reduced dramatically to less than half. At present time, there are around 320 farmers involved in the cultivation and marketing of mushrooms in Peninsular Malaysia. Most of them are small farmers who produce 50-500 kg of fresh mushrooms.

State	Number of growers	Production (kg/day)
Selangor	48	7830
Johor	67	7800
Kelantan	22	4565
Negeri Sembilan	30	1036
Pahang	50	550
Kedah	9	455
Pulau Pinang	13	390
Melaka	16	360
Perak	30	345
Terengganu	29	265
Perlis	4	210
Total	318	23,806

Table 2.2.2 Table of mushrooms growers by state

Source: Department of Agriculture Malaysia, 2012.

More than 300 farmers and companies cultivated mushrooms in Peninsular Malaysia with a total production of approximately 24,000 kg per day in 2012. Out of the total farmers and companies, only 6 of the companies produce a large scale of over 500 kg per day while around 60 companies produce in moderate scale of about 50-500 kg per day. The remaining farmers are considered as small-scale growers. Small scale growers make up about 80% of the total population of mushrooms growers, medium scale at 17% and big scale are about 3%.

2.3 *Pleurotus florida*

Pleurotus spp represent the third largest group of cultivated mushrooms in the world. They are mostly grown on a variety of plant residues and they have been found nutritionally and gastronomically important (Cohen *et al.* 2002). Among the traditionally cultivated and potentially new mushrooms species is the white oyster mushrooms (*Pleurotus florida*). *P.florida* are known to be had extraordinary opportunities for cultivation and consumption.

Pleurotus species belong to the family of *Pleurotaceae* family which is the member of *Basidiomycota* phylum. There are white saprobe and facultative parasitic species in the genus that degrade the polymers of lignin and cellulose. *P.florida* varieties can be found on pastures, meadows, gardens and seldom in grassy forest and hilly areas. This species is mainly associated with the members of the *Apiaceae (Umbelliferae)* plant family in its natural habitat (Ewald, 2007).

Kingdom	Fungi
Phylum	*Basidiomycota*
Class	*Agaricomycetes*
Order	*Agaricales*
Family	*Pleurotaceae*
Genus	*Pleurotus*
Species	*P.florida*

Table 2.3.1 Taxonomy description of *Pleurotus florida*

This mushroom also carries statins, which are disease fighting compounds. The particular compound found in *Pleurotus* spp, is called Lovastatin. It is a compound that helps to clear cholesterol from the circulatory system. Once the blood is free of cholesterol, blood circulation is enhanced and the body feels healthier. A research done on mice indicated that *P. florida* has the effect of controlling insulin in the body (Sullivan, 2002).

2.4 Cultivation of *Pleurotus florida*

Different strains of oyster mushroom response differently to different substrates, supplement, supplementation amount and environmental factors in the aspects of mycelium run, average yield and quality. It can easily and successfully be cultivated on wheat and rice straw, cotton waste and sawdust. Study done by Baysal (2003), cultivation of oyster mushroom can be done on waste paper substrate with proper amount of supplement added. The temperature range required for successful cultivation is 20 to 25^0c for fruiting body development.

The growing environment for the mushrooms should be brought as close as possible to the natural environment for the successful fruiting bodies development. The effects of environmental factors such as light intensity, temperature, humidity (Cheng and Han, 1997) indicate that light intensity produced no significant effect on the growth whereas both temperature and humidity had a significant effect on growth and formation of fruit bodies of *pleurotus* species. Generally, there is no fruiting bodies formation under humidity of less than 65% and there is an abnormal resulted in a saturated atmosphere. According to Parjimo and Andoko (2012), the growth of *pleurotus* species is stunted when being cultivated in an environment with the relative humidity less than 60%. High relative humidity also will disturb the growth of mushroom and infection from diseases and pests are easily to be occurred.

According to Won (2004), carbon dioxide concentration play an important role in the cultivation of mushrooms. The optimum carbon dioxide concentration for successful cultivation of oyster mushroom had been recorded to be about approximately 15-20%. Higher carbon dioxide concentration may result in the failure of the mycelium to be formed completely and the formation of the cap or pileus of the mushroom deteriorate. Good aeration system should be installed to control the rate of carbon dioxide concentration in controlled environment.

2.4.1 Life cycle of mushrooms

The life cycle of typical mushrooms consist of 3 stages. The first stage occur when there is a combination of basidium to produce zygote from the diploid cell. The first stage occur at the under the surface of the cap or pileus of the mushroom which is full of sexual reproductive gills-like structure. The second stage in the life cycle of typical mushroom occur when the zygote undergoes the process of meiosis and basidiospores will form sterigmata. The last stage of the life cycle of mushroom will occur when basidiospore will be out from basidia structure to germinate into hypae. The formation of hypae will produce mycelium. Hypae will continue to germinate to produce more mycelium. The mass of mycelia will form structures called as basidiocarp which is the fruiting bodies (Alan, 2007). Generally, the life cycle of mushrooms start from the formation of mycelia which produced from the spores. Table 2.4.1 showed the main stages of the mushroom growth.

Stages	Explanation/description of stages
I (Week 1-2)	-Mycelium growth -Formation of hypae to form mycelia
II (Week 3)	-Formation of pinhead -Light intensity and humidity is important
III (Week 4)	-Formation of two main structures which are cap (pileus) and stem/stalk -Cap (pileus) completely formed

Table 2.4.1 Stages of mushroom growth

Source: Abdul Rahman, 2008.

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