

UTILIZATION OF COCONUT WATER AS INDUCING SUBSTANCE IN THE CULTIVATION OF *Pleurotus ostreatus*

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ABSTRACT

Cultivation of edible mushrooms has been evaluated as an effective way for increasing income of people in developing countries. *Pleurotus ostreatus*, well known as an oyster mushroom is the most popular edible mushroom in Indonesia. They usually is cultivated using a sawdust as a main substrate. The aim of this research is to reveal the addition of coconut water, the byproduct of traditionally coconut process as an inducing substance in *P. ostreatus* cultivation. The results shown that the addition 25% (v/v) of coconut water in substrate of *P. ostreatus* induce the growth of mycelium. On the other hand, the addition of 50% (v/v) of coconut water affect in the day of fruit body formation become faster as well as increase the number of pileus (stalk) and the weight of fruit body.

Keywords: Coconut water, *Pleurotus ostreatus* cultivation

INTRODUCTION

Edible mushroom cultivation provides an economical benefit of agricultural industry activity that has markedly developed in the word in the past few decades. *Pleurotus ostreatus*, well known as an oyster mushroom is the third largest commercially produced mushrooms in the world since it is a prospective source of cheap valuable food protein with the ability to effectively bioconvert various lignocellulosic materials (Kues, U and Liu, Y. U. 2000; Saskiawan, I et al. 2016, Islam W and Raiz, A. 2017). Mandel et al. 2005 reported that *P. ostreatus* was successfully cultivated using four different waste materials, unsorted office paper, cardboard, plant fibres (*Bromus fasciculatus*) and white sawdust as a

substrate from more feasible and cheap recyclable residue.

Furthermore, mushroom have become attractive as a functional food and as a source for the development of drugs and nutraceutical. It is because mushroom has antioxidant, antitumor, and antimicrobial properties as well as becoming more important in our diet due to their nutritional value. It related to the high protein and low fat/energy contents (Kumar, 2015). Recently, cultivation of *P. ostreatus* in Indonesia has been increasing quickly. The reasons are very simple of its cultivation technology as well as the abundantly of substrate availability for growing medium.

The coconut (*Cocos nucifera* L.) is an important fruit tree in the tropical regions

and the fruit can be made into a variety of foods, beverages and its derivative. Every part of the plant is useful in human life. The leaf can be used as a traditional food wrap. The trunk and wood is a good for building material. The fruit have the most economical value. Its mesocarp, called the husk is processed into rope, carpets, geotextiles and growing media. On the other hand, the hard brown shell (endocarp) can be processed into high quality activated charcoal. The inner part or the nut (endosperm) is divided into two edible portion: a white kernel and a clear liquid: coconut water (Prades, et al. 2012). In Indonesia the coconut water is quite abundant since the using of it in beverage industry has not been maximized. The aim of this study is to reveal the utilization of coconut water as an inducing substance in formation of fruiting body in *P. ostreatus* cultivation.

RESEARCH METHODS

Strain of *P. ostreatus*

P. ostreatus strain Cibinong was used in this study and deposited in the Indonesian Culture Collection (Ina CC), Research Center for Biology, Indonesian Institute of Sciences. It is maintained under paraffin oil and re-cultured on a potato dextrose agar (PDA) slant before using in this study.

Spawn Preparation

The main substrate for the mother spawn is a sorghum in good quality which contain few broken kernels, little debris and be recently harvested. The grain was soaked for overnight and the floated one was not used. It was drained and cooked in water for 10 to 15 minutes. It was drained again and added with 1% (w/w) of ground limestone (CaCO_3). Then, the substrate was put into a spawn bottle and sterilized at 121 °C for 30 min. After sterilization, each bottle was inoculated with 6 mm square mycelium of *P. ostreatus* growth on PDA in petri

disk. Then, it was incubated at 30 °C in the dark for 14 days.

The mother spawn is used a source for inoculation into final spawn. The main substrate of final spawn are sawdust (73%), rice husk (20%), corn flour (5%), limestone (1%) and gypsum (CaSO_4) (1%). All substrate was mixed and add 65% of water. It was then put into polypropylene bag and cover with cotton. One day after sterilization, it was inoculated with 30 g of mother spawn an incubated at 30 °C in the dark for 14 days.

Substrate for Growing *P. ostreatus* preparation.

The main content of substrate in *P. ostreatus* cultivation is sawdust with the addition of CaCO_3 (2%), CaSO_4 (1%), rice bran (15%) and corn flour (4%). First, the sawdust and CaCO_3 was composted for 5 days. Then, the other materials were added and mixed properly. Tap water containing various concentrations of 0%, 25%, 50%, 75%, and 100% of coconut water was added as a treatment in this study to obtain a moisture content of 70%. Polypropylene bags (15x35 cm) were filled with 1.0 kg of substrate, covered with cotton, and sterilized at 100 °C for 2 hours and allow to cool down to the desired inoculation temperature. The next day after sterilization, the bags were inoculated with the final spawn at a rate of 5% of the substrate weight and incubated in an incubation room maintained at 28 + 5 °C at a relative humidity of 70-75 + 5%. After the substrate was fully colonized with mycelium, the bags were replaced in a growing room and incubated at 25 + 5 °C at relative humidity of 80-85 + 5 %.

Harvesting and Analysis.

In the growing room, the bags were opened. After the opening of the bags, 7-10 days later, the primordial were formed. The fruit body was harvested by clasping the stipe and gently twisting and pulling the fruiting body from the

substrate. Parameters which are determined include the growth of mycelium, the first formation of fruiting body, the number of fruiting body, the weight of fruiting body, and the interval time of fruiting body formation. Data were analyzed using the analysis of variance (Anova) procedure by SPSS 16 and means were separated using Duncan's multiple range test (DMRT) at $p < 0.05$.

RESULTS AND DISCUSSION

The growth of *P. ostreatus* cultivated using a bag medium system, can be determined by measuring the length of mycelium that spread from the top to the bottom of bag. The results showed that the concentration of coconut water of 100% was the most favorable for mycelium growth. It was 40.1 mm after 3 days, 125.5 mm after 15 days, and 201.1 mm after 27 days (Table 1). Statistically, they only significantly different on 3 days and 15 days after inoculation with spawn. In this study, the harvesting of fruit body was done until the second fruit body production in a bag.

Table 1. The growth of mycelium of *P. ostreatus* on the substrates with addition of various concentration of coconut water

Concentration of coconut water	The growth of mycelia after inoculation (mm)		
	3 days	15 days	27 days
0%	38.0 ^a	123.1 ^c	192.6 ^{bc}
25%	38.6 ^a	119.8 ^b	194.3 ^{abc}
50%	38.6 ^a	115.2 ^a	186.8 ^{ab}
75%	38.8 ^a	113.7 ^a	179.7 ^a
100%	40.1^b	125.5^d	201.1^c

Explanation: Values with different superscripts in the same row are significantly different at $p < 0.05$ based on DMRT

The addition of coconut water of 25% gives the fastest of fruit body formation of 24.56 days as well as the shortest period of fruit body formation of 76.60 days (Table 2). Both of the first fruit body formation and period of fruit body formation were not significantly different compare to the addition of other concentration of coconut water. However, it was significantly different compare to the control (0% of coconut water). Table 2 also shown the number and the total weigh of fruit body. It was the most number of pileus of 35.24 and the heaviest of 273.92 gr of fruit body as a result of the addition 25% and 50% of coconut water.

The *P. ostreatus* was characterized by the production of fruit bodies with an eccentric stipe and a wide pileus shaped like an oyster shell, with the widest portion of the pileus being away from the stipe. Figure 1. shown the character of fruit body of *P. ostreatus* affected by the addition of coconut water onto its substrate. The number of pileus in substrate without addition of coconut water is less than that of pileus in substrate added with coconut water. Consequently, the size of pileus in substrate without addition of coconut water was also larger than that of without addition of coconut water..

Tabel 2. The first fruit body formation, the period of fruit body production, the number of pileus and the weight of fruit body on the substrates with addition of various concentration of coconut water

Concentration of coconut water	First Fruit Body Formation (days)	Period of Fruit Body Production (days)	Number of pileus	Weight of Fruit Body (g)
0 %	36.72 ^b	90.20 ^b	22.48 ^a	156.64 ^a
25 %	24.56^a	76.60^a	35.12 ^b	245.96 ^b
50%	28.24 ^a	81.44 ^a	35.24^b	273.92^c
75%	27.52 ^a	80.20 ^a	32.00 ^b	227.80 ^b
100 %	25.80 ^a	79.04 ^a	34.56 ^b	270.52 ^c

*Values with different superscripts in the same row are significantly different at $p < 0.05$ based on DMRT

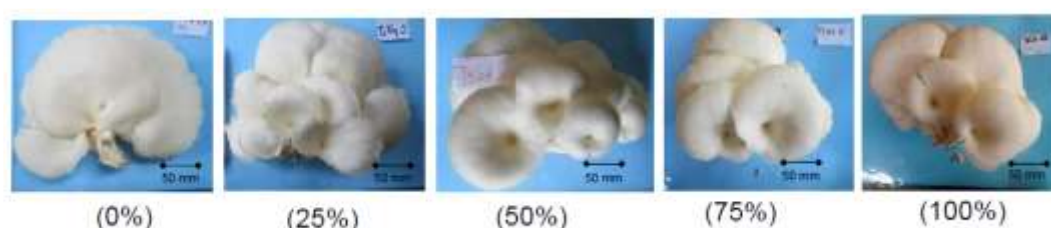


Figure 1. The fruiting body of *P. ostreatus* in substrate with addition of various concentration of coconut water

Mycelium (singular: mycelia) is a vegetative phase of mushroom that totally obtains nutrients from a substrate. Regarding measuring the length of mycelium as the determination of the growth of mushroom, it showed that the results of this study is similar as the study of De Leon et al (2013). The study reported the suitability of coconut water as medium for *Lentinus* spp. and *Polyporus gramocephalus*. Furthermore, Jacob et al. (2015) also reported the coconut water gelatin supported the mycelial growth of the *P. citrinopileatus*, *P. djamor* and *P. salmoneostramineus* as indicated by the luxuriant mycelial growth. The ability of the coconut water to promote mycelial growth of *P. ostreatus* is attributed to its nutritional content. Fresh coconut water is hygienic and nourishing. As a fluid endosperm of the coconut fruit, it contains minerals as well as other essential nutrients for the induction of morphogenesis such as inducing fungal cells to divide and grow rapidly (Magday et. al. 2014).

Furthermore, coconut water is widely used in the plant tissue culture industry. The extensive use of coconut water as a growth-promoting component in tissue culture medium formulation can be traced back to more than half a century ago. From a scientific viewpoint, the addition of coconut water to the medium is rather unsatisfactory, as it precludes the possibility for investigating the effects of individual components of the medium with any degree of accuracy. The question of which components cause the growth stimulation arose immediately. Besides its nutritional role, coconut water also appears to have growth regulatory properties, e.g., cytokinin-type activity which are a class of phytohormones (Yong et al. 2009).

The fruit body formation of *Pleurotus* spp. is related to both intrinsic and extrinsic factors. The intrinsic factors include composition of substrates, sources of nitrogen, ratio of carbon to nitrogen (C/N), pH, moisture, minerals, vitamin, particle size and levels of

spawning. Furthermore, the extrinsic factors include humidity, temperature, luminosity, and air composition (Belletini et al. 2016). Growth parameters of temperature and humidity for *P. ostreatus* is a 24 °C, 85-95%; 10-15.6 °C, 95-100% and 10-21 °C, 85-90% for spawn running, primordial formation and fruit body development respectively. (Stamets, 2000). However, in this study the measurement of microclimate showed 28-30 °C, 70-80% for spawn running and 27-29 °C, 80-90% for primordial formation and fruit body development in the mushroom house. However, Islam et al. 2016 reported that mycelial growth of all oyster mushroom can take place between 20 °C and 30 °C. However, for fruiting, different species have different temperature requirements. *Pleurotus* spp. grows in wide range of temperature from 15 to 30 °C.

Furthermore, the addition of coconut water was also affect in the time of fruit body formation. For the mushroom growers, the faster of fruit body formation, the more profit will be obtained. Mushroom growers can immediately change the spent mushroom substrate with the new one. Consequently, they will get faster capital turnover.

CONCLUSIONS

In the cultivation of *P. ostreatus*, addition of coconut water in the substrate affect in the growth of mycelium in the vegetative phase. It also make the day of fruit body formation become faster, number of pileus as well as the weight of fruit body. The addition of 50% (v/v) of coconut water give the heaviest of fruiting body.

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REFERENCES

- Belletini MB., Fiorda FA., Maieves HA., Teixeira GL., Avila S., Hornung, PS., Junior AM., and Ribani RH. 2019. Factor affecting mushroom *Pleurotus* spp. Saudi Journal of Biological Sciences, 26: 633-646
- De Leon AM., Reyes RG., and Dela Cruz TEE. 2013. *Lentinus squarrosulus* and *Polyporus gramocephalus*: newly domesticate, wild edible macrofungi from the Philippines. The Philippine Agricultural Scientist, 96 (4): 411-418
- Islam MDT., Zakaria Z., Hamidin N., and Mohdishak MAB. 2016. Effects of natural environment and increased ranges of humidity for indoor cultivation of grey oyster (*Pleurotus pulmonarius*) mushroom in Malaysia. International Journal of Advances in Science Engineering and Technology, 4(2):121-127
- Islam W. and Riaz A. 2017. Yield and Biological Efficiency of *Pleurotus ostreatus* (Jacq. Fr.) cultivated upon various weeds and agricultural wastes. Pakistan Journal Weed Science Research, 23(3): 271-279
- Jacob JKS., Kalaw SPK., and Reyes RG. 2015. Mycelial growth performance of three species of *Pleurotus* on Coconut Water Gelatin. Current Research in Environmental and Applied Mycology, 5 (3): 263-268
- Kues U., and Liu Y. 2000. Fruiting body production in Basidiomycetes. Applied Microbiology Biotechnology, 54: 141-152
- Kumar K. 2015. Role of edible mushrooms as functional foods-A review. South Asian Journal of Food Technology and Environment, 1 (3&4): 211 -218
- Magday Jr. JC., Bungihan ME., and Dulay RMR. 2014. Optimization of mycelial growth and cultivation of

- fruiting body of Philippine wild strain of *Ganoderma lucidum*. *Current Research in Environmental & Applied Mycology*, 4(2), 162-172
- Mandel QA., Al-Lait AA., and Mohamed, SA. 2005. Cultivation of oyster mushrooms (*Pleurotus* spp.) on various lignocellulosic wastes. *World Journal of Microbiology & Biotechnology*, 21:601–607
- Prades A., Dornier M., Diop N., and Pain JP. 2012. Coconut water uses, composition and properties: a review. *Fruits*, 67: 87-107
- Saskiawan I., Hasanah N., and Shimomura N. 2016. Cultivation of *Pleurotus ostreatus* using sorghum-supplemented spawn on various substrates. *Mushroom Science and Biotechnology*, 23 (4): 179-182
- Stamets P. 2000. Growing gourmet and medicinal mushrooms. Ten Speed Press. Box 7123 Berkeley, California. USA page: 313
- Yong JWH., Ge L., Ng YF., and Tan SN. 2009. The chemical composition and biological properties of coconut (*Cocos nucifera* L.) Water. *Molecules*, 14: 5144-5164.