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Waste, Tubers and Seeds from Peat Land as Alternative Media for Fungal Growth Media

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ABSTRACT

Research to investigated the use of alternative media from waste, tubers and seeds from peatlands as alternative media for fungal growth has been carried out for 3 months and the last research was conducted in November 2024. In the first stage the materials used are tuber and peel from *Solanum tuberosum*, *Manihot esculenta*, *Pachyrhizus erosus*, *Ipomea batatas* (yellow, orange and purple), *Colocasia esculenta* var kujang enyuh and kujang malahoi, also peel from *Cucubita moschata*, *Ananas comosus* and *Carica papaya* used to growth *Sclerotium rolfsii* and *Capsicum capsici*. In the second stage, the materials used were seed extract and powder of *Artocarpus champeden, Durio zibethinus* and *Terminalia catappa* and *S. tuberosum* extract used to growth *Pleurotus ostreatus, Trichoderma koningii* and *Gliocladium virens*. The results showed that are good media for the growth of *S. rolfsii* and *C. capsisi* are pineapple peel, papaya peel, pumpkin peel, *C. esculenta* var kujang enyuh and kujang malahoi tuber, *P. erosus* tuber, tubers and peel of purple sweet potato. Good media for the growth of *P. ostreatus, T. koningii* and *G. virens* are seed extract and powder from *A. champeden, D. zibethinus,* and *T. catappa* also and *Solanum tuberosum* extract. This study reported that the media from tubers and peel from *C. esculenta* var kujang enyuh and kujang malahoi, peel from cassava, sweet potato, potato, pineapple fruit, pumpkin fruit, papaya fruit, also seeds from *T. catappa* were new findings for fungal growth media that had not been reported before by other researchers.

Key words: peel, waste, tuber, seed, media, growth, fungi

INTRODUCTION

Now days, the media for growth of microorganisms use commercial synthetic media like nutrient broth (NB) for bacteria and Potato Dextrose Agar (PDA) for fungal growth where the prices of commercial synthetic media is getting more expensive every time. Alternative media for bacterial growth that are cheaper than organic materials and have quality that is not much inferior to commercial NB media include coconut water media, green bean sprout waste water, tofu waste water, rice washing water, fish bone stew, chicken bone stew, boiled shrimp shells, boiled snail meat, and boiled fish intestines (Nion *et al*., 2016).

The media for fungal propagation that is most widely used at present is PDA media with the main ingredient coming from potato tubers (Laurie *et al*., 2015) but there are still few res earches on the use of alternative media from other organic materials, especially plants from peat land. Based on its composition, PDA is included in semi-synthetic media because it is composed of natural ingredients (potatoes) and synthetic materials (dextrose and agar). Potato is a source of carbohydrates, vitamins and energy, dextrose is a source of sugar and energy, besides that the agar

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component functions to condense the PDA medium. Each of the three components is necessary for the growth and proliferation of microorganisms, especially fungi. Physical and nutritional factors affect the growth of fungi (Abdulla, 2018).

Starch-producing tubers that have been identified as alternative media for fungal growth are cassava flour (Kwoseh *et al*., 2012; Askari, 2018), Dioscorea alata (Sawiphak *et al*., 2021), sweet potato and taro (Wongjiratthiti & Yottakot 2017), carrots and sweet potatoes (Rezekikasari & Harianto, 2019), while for bacteria growth media are Pachyrrhizus erosus tubers and Vigna radiate media (Rizki & Syahnita, 2019). Alternative media from seeds and nuts that are known are cowpeas, green beans, black soybeans (Ravimannan *et al*., 2014), Artocarpus integer seed, jackfruit seed, durian seed (Betharia, 2017), and alternative media from fruit still little is known (Rezekikasari & Harianto, 2019). There have never been examined the waste from fruit peel as an alternative medium for fungal growth. During the fruiting season, the seeds of chempedak (*Artocarpus integer*), durian (*Durio zibethinus*) and Indian amond (*Terminalia cattapa*) are very abundant and often become waste that has never been utilized. Therefore, some parts of this research seeked to utilize these wastes as an alternative medium for fungal growth.

MATERIALS AND METHODS

This study used a completely randomized design (CRD) using various types of media as treatments (Table 1). The different among treatments were examined based on Analysis of Variance (ANOVA). If they were significantly different, they were analyzed using Tukey's honestly significance difference (HSD).

There were 14 types of alternative media used to growth Sclerotium rolfsii, Capsicum capsici, Pleurotus ostreatus, Trichoderma koningii and Gliocladium virens fungi.

Preparation of Solid or Liquid Media from Fresh Extracts Media

To make 1 liter of media, the way is as follows: each ingredient for alternative media is weighed 200 g, washed and then cut into small cubes of about 1 cm in size. The ingredients are put into the pot, added 1000 mL distilled water, 15 g agar (if make liquid media not used agar) and 20 g sugar and then boiled until soft. The extract was filtered and put into a beaker glass, after which it was sterilized using an autoclave at 121 °C for 15 minutes. The media is ready to be poured into the petridish to test fungal growth. If test used liquid media, media poured 15 mL into beaker glass/bottle glass.

Preparation of Solid or Liquid Special Powder Media from Chempedak Seed, Durian Seed, Indian Almond Seed and Potato Tubers

To make one liter of media, the method is as follows: each ingredient of chempedak seeds, durian seed, Indian almond seed and potato tubers were cut thinly and dried in the sun light until dry. The dry ingredients were pulverized using a blender or mortar and pestle until a fine flour consistency was achieved. A total of 20 g of flour was added to a pot containing 1000 mL of distilled water, 15 g of agar (excluded if liquid media was prepared), and 20 g of sugar. The mixture was then boiled until a homogeneous solution was obtained. The resulting medium was transferred into a beaker glass and sterilized using an autoclave at 121°C for 15 minutes. After sterilization, the medium was ready to be poured into Petri dishes for fungal growth testing. For tests using liquid media, 15 mL of the medium was dispensed into a beaker glass or a bottle.

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Table 1. Alternative media used in this research

No.	Latin name	English name	Indonesia name	Dayak Ngaju Name	Source	Organ used
1	*Solanum tuberosum*	Potato	Kentang	Kantang	Mineral soil, Java island	Tuber and peel
2	*Manihot esculenta*	Cassava	Singkong	Jawau	Peat soil, Kalampangan	Tuber and peel
3	*Pachyrhizus erosus*	Yam bean	Bengkoang	Bengkuang	Peat soil, Kalampangan	Tuber and peel
4	*Ipomea batatas*	Orange Sweet potato	Ubi jalar orange	Gambili bahenda batue	Peat soil, Kalampangan	Tuber and peel
5	*Ipomea batatas*	Yellow Sweet potato	Ubi jalar kuning	Gambili bahenda	Peat soil, Kalampangan	Tuber and peel
6	*Ipomea batatas*	Purple Sweet potato	Ubi jalar ungu	Gambili ungu	Peat soil, Kalampangan	Tuber and peel
7	*Colocasia esculenta* var Kujang Malahoi	Taro var Kujang Malahoi	Talas var Kujang Malahoi	Kujang Malahoi	Peat soil, Pilang	Tuber and peel
8	*Colocasia esculenta* var Kujang Enyuh	Taro var Kujang Enyuh	Talas var Kujang Enyuh	Kujang Enyuh	Peat soil, Pilang	Tuber and peel
9	*Cucubita moschata*	Pumpkin	Labu kuning	Bajawa	Peat soil, Kalampangan	Peel
10	*Ananas comosus*	Pineapple	Nenas	Kanas	Peat soil, Kalampangan	Peel
11	*Carica papaya*	Papaya	Pepaya	Kastela	Peat soil, Kalampangan	Peel
12	*Artocarpus integer*	Chempedak	Cempedak	Mangkahai	Peat soil, Jabiren	Seed
13	*Durio zibethinus*	Durian	Durian	Dahuyan	Mineral soil, Kasongan	Seed
14	*Terminalia catappa*	Indian almond	Ketapang	Ketapang	Peat soil, Jekan Raya sub-district	Seed

Fungal Growth Test on Solid and Liquid Media

For solid media, the sterilized alternative medium was poured into petri dishes and allowed to solidify at room temperature. A 0.5 cm diameter plug was cut from the fungal isolate using a cork borer, then transferred to the center of the medium using a sterile inoculating needle. The inoculated media were incubated at room temperature, and fungal growth was observed daily for up to 4 days after inoculation (dai).

For liquid media, a 0.5 cm diameter plug was excised from the fungal isolate using a cork borer and transferred into the liquid medium using a sterile inoculating needle. The inoculated media were incubated at room temperature, and fungal growth was observed for up to 4 weeks after inoculation.

RESULTS

Fungal Growth on Fresh Extract Media from Tubers and Peels Waste

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The results of the study indicated waste from tuber and fruit peels can be used as an alternative medium for fungal growth.

Table 2. Fungal growth on tubers and peel waste media

Treatment	Diameter (cm) of *S. rolfsii at* day					Diameter (cm) of *C. capsici at* day					
	3	4	5	6	7	3	4	5	6	7	8
Control	6,8^{b}	9^{c}	9^{c}	9^{b}	9	3,7	5,75^{b}	6,6^{e}	7,05^{cde}	8^{c}	9
S. tuberosum -tuber	6,65^{b}	8^{bc}	9^{c}	9^{b}	9	1,6	3^{ab}	4^{ab}	4,9^{abc}	6,75^{abc}	8^{abc}
S. tuberosum -peel	6,25^{b}	8^{bc}	9^{c}	9^{b}	9	1,5	4^{ab}	4,5^{abc}	4,8^{ab}	5,75^{ab}	6,9^{ab}
M. esculenta -tuber	6^{b}	8^{bc}	9^{c}	9^{b}	9	3,5	4,5^{ab}	6de	6,65^{cde}	7,4^{bc}	8^{abc}
M. esculenta -peel	6^{b}	8,25^{bc}	9^{c}	9^{b}	9	2,9	3,5^{ab}	5,5^{cde}	6,25^{cde}	6,65	7,1^{ab}
Pachyrhizus erosus -tuber	6,25^{b}	8^{bc}	9^{c}	9^{b}	9	3,8	4,5^{ab}	5^{bcd}	6,1^{bcde}	7,25^{bc}	9^{c}
Pachyrhizus erosus -peel	2,75^{a}	3,75^{a}	6,25^{a}	6,65^{a}	9	1,7	2,4^{a}	4,5^{a}	4,15^{a}	5,25	6,5^{a}
I. batatas -orange-tuber	6^{b}	6,5^{b}	9^{c}	9^{b}	9	2,1	4^{ab}	4,5^{abc}	6^{bcd}	6,9^{abc}	8^{abc}
I. batatas -orange-peel	6^{b}	6,75^{bc}	7,9^{b}	9^{b}	9	3	4^{ab}	5^{bcd}	5,9^{bcd}	6,75^{abc}	7,5^{abc}
I. batatas -yellow-tuber	6,05^{b}	8^{bc}	9^{c}	9^{b}	9	4	4,5^{ab}	5,75^{cde}	6,9^{cde}	7,65^{c}	8,4^{bc}
I. batatas -yellow-peel	6,05^{b}	8^{bc}	9^{c}	9^{b}	9	3,5	4,4^{ab}	5,75^{cde}	7,15^{de}	7,75^{c}	8,25^{bc}
I. batatas -purple-tuber	6^{b}	9^{c}	9^{c}	9^{b}	9	4	5^{ab}	6,5^{e}	7,35^{e}	8^{c}	8,25^{bc}
I. batatas -purple-peel	6,65^{b}	9^{c}	9^{c}	9^{b}	9	4	5^{ab}	5,5^{cde}	6,9^{cde}	7,5^{c}	8,5^{bc}
Colocasia esculenta var Kujang Malahoi-tuber	6^{b}	8,5^{bc}	9^{c}	9^{b}	9	3,4	4,4^{ab}	5^{bcd}	6,5^{cde}	7,9^{c}	9^{c}
Colocasia esculenta var Kujang Malahoi-peel	6,25^{b}	8,5^{bc}	9^{c}	9^{b}	9	4	4,5^{ab}	6de	6,9^{cde}	7,5^{c}	8,5^{bc}
Colocasia esculenta var Kujang Enyuh-tuber	5,8^{b}	8^{bc}	9^{c}	9^{b}	9	3,9	4,5^{ab}	5^{bcd}	6,1^{bcde}	7,4^{bc}	9^{c}
Colocasia esculenta var Kujang Enyuh-peel	5,7^{b}	7,5^{bc}	8^{b}	9^{b}	9	3,5	4^{ab}	6de	5,75^{bc}	6,9^{abc}	8^{abc}
C. moschata -peel	5,7^{b}	8^{bc}	9^{c}	9^{b}	9	3,25	4,25^{ab}	6de	6,9^{cde}	8^{c}	9^{c}
A. comosus -peel	6,5^{b}	9^{c}	9^{c}	9^{b}	9	2,1	4,5^{ab}	6de	6,45^{cde}	7,8^{c}	9^{c}
C. papaya -peel	6,25^{b}	9^{c}	9^{c}	9^{b}	9	2,5	3^{ab}	4,75^{bcd}	6^{bcd}	7,9^{c}	9^{c}

Note: Means followed by the same letter within a column are not significantly different according to Tukey's honestly significance difference (HSD), 1%

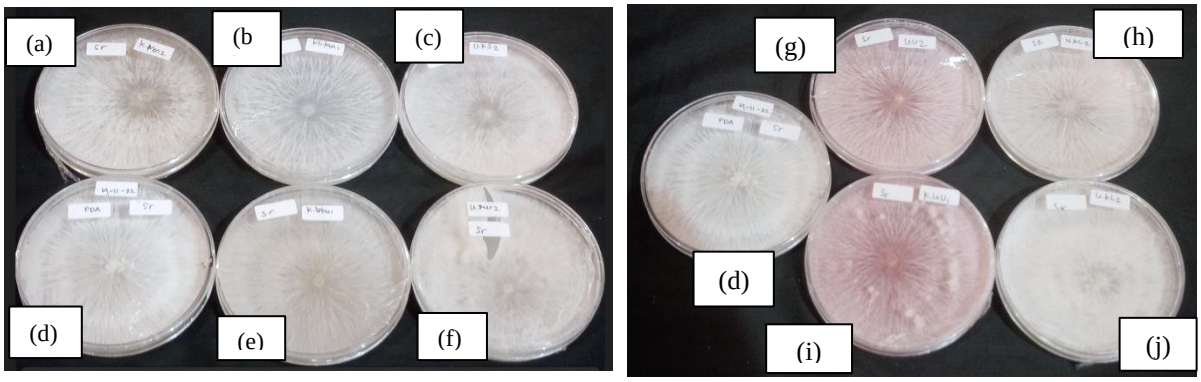

Figure 1. *S. rolfsii* good growth on the alternative media (a). pineapple peel, (b). yam bean peel, (c) Cassava tuber, (d) PDA-control, (e) pumpkin peel, (f). orange sweet potato tuber, (g). purple sweet potato tuber, (h). taro var. kujang malahoi peel, (i). purple sweet potato peel, (j). taro var. kujang malahoi tuber.

Fungal Growth on Fresh Extract Media from Seed

The fastest growing fungus was T. koningii, followed by G. virens, while the growth of was rather slow (Table 4; Figures 2 & 3). This is presumably because the nutritional needs of each fungi were different and the growth rate was also different. Fungal growth was generally faster in fresh extract seed media than in powder seed.

On solid media, on the third day, statistically there was no difference in the speed of growth of all fungi compared to the control media except for fresh extract and powder durian seed but after 4 days (slower). Although statistically not significantly different, in fact, the fastest medium for the growth of the three fungi is Indian almond seed media.

Fungal growth was faster on liquid media than on solid media. At liquid media aged 4 days there was an interaction between the type of media and the type of fungus, where the best medium was Indian almond seed powder on the growth of T. koningii and G. virens.

Fungal grow in both solid and liquid media, in general the fastest growth is from Indian Amond seeds, chempedak and followed by durian seeds (Figures 2 & 3). Indian almond seeds have good nutrition for microbial growth, especially the sugar content (2.38 to 3.16 g), protein (23 to 25 g) and fat (60 g) are higher than the nutrients in potato tubers (Weerawatanakorn *et al*., 2015). This probably the reason why the growths of all fungi on Indian almond seed media were higher than the media from other seeds (Table 3).

DISCUSSION

The peel waste of *A. comosus* and *C. papaya* grows very well on both types of fungi, both *S. rolfsii* and *C. capsici*, this is due to the relatively high nutrition in the peel of *A. comosus* and *C. papaya*, especially containing 29.2 to 35.8 g of sugar and 1.78 g respectively higher than potato tubers (1.2 g), as well as fat and protein content. Likewise, the higher sugar content was found in the tubers of *I. batatas*-puple (5 g*), C. esculanta* (0.2 to 2 g) and *P. erosus* (1.0 to 3.4 g) (Aryanti *et al*., 2017; Giri, 2019; Jaiswal et al., 2022; Kamoona *et al*., 2015; Kim *et al*., 2020; Kusumaningsih et al, 2022; Sorensen, 1996; Temesgen & Retta, 2015 ; Yuliansar et al., 2020). Fungi need nutrients such as a carbon skeleton (derived from carbohydrates and sugar), water, oxygen, temperature and other factors for growth and energy production for example in *Aspergillus niger* it has been shown that sugar is the only carbon source (Hamad *et al*., 2014).

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The pineapple peel contained an appreciable amount of alcohol-insoluble solids (AIS), and water-insoluble solids (WIS), which primarily consisted of hemicellulose substance and pectic, also cellulose, insoluble fiber-rich fraction (FRF) (41.8 - 48.0 g/100 g) including insoluble dietary fiber (IDF), These fractions also contained notable proportions of lignin (60.7 - 65.8 g/100 g). In terms of sugar composition, these insoluble fiber-rich fraction (FRF) contained a considerable amount of cellulosic glucose (8.33 - 10.7 g/100 g), followed by xylose (7.83 - 9.23 g/100 g) and uronic acid (6.83 - 8.26 g/100 g). It can thus be deduced that these fractions contained certain polysaccharides, such as cellulose, hemicellulose (xylans or xyloglucan), and pectic substances, as major constituents (Huang *et al*., 2011).

This supports the results of research that alternative media containing sugar which is almost equivalent to potatoes value or higher than potatoes can be a good alternative medium for fungi growth. Pineapple peel waste represents around 35% of the whole fruit mass (Huang *et al*., 2011), meaning that the media from pineapple peel waste for fungal growth media has great potential to be utilized.

Chempedak seeds are a source of carbohydrates (36.7 g/100 g), which is three times the amount of wheat seeds. The protein of cempedak seeds (4.2 g/100 g) almost exceeds half of the content of wheat, with energy (165 kcal/100 g), has a good source of minerals and so it is not surprising that it can be a source of alternative media for microbes (Ansari et al, 2010).

The carbohydrate content of durian seeds is not much different from the nutrition of potato tubers but has a higher protein value than potatoes. Other mineral content is sufficient for microbial growth, but durian seeds contain a lot of mucilage, the main components of which are phosphatides, proteins, carbohydrates and water, which are odor less and tasteless, soluble in water, and have high water binding capacity (Amid and Mirhosseini, 2012).). That is why the growth of mushrooms in media from durian seeds is slower than other seeds because the seeds contain mucus so that the mushrooms are less free to grow.

CONCLUSSION

Good media for the growth of *S. rolfsii* and *C. capsisi* are pineapple peel, papaya peel, pumpkin peel, *C. esculenta* var kujang enyuh and kujang malahoi tuber, *P. erosus* tuber, tubers and peel of purple sweet potato. Good media for the growth of *P. ostreatus*, *T. koningii* and *G. virens* were seed extract and powder from *A. champeden*, *D. zibethinus*, and *T. catappa* also and *Solanum tuberosum* extract.

This study proved that the media from tubers and peel of *C. esculenta* var kujang enyuh and kujang malahoi, peels (cassava, sweet potato, potato, pineapple fruit, pumpkin fruit, papaya fruit), also seeds of *T. catappa* were new findings for fungal growth media that had not been reported before by other researchers.

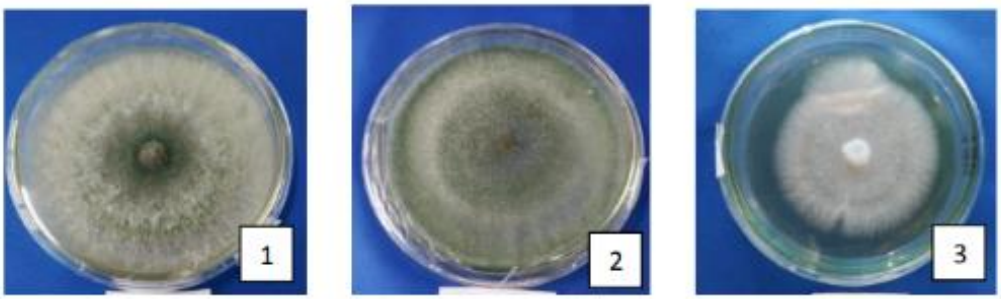

Figure. 2. Indian almond seed media (solid media) is good media for growth (1). *T. koningii*, (2). *G. virens*, and (3). *P. ostreatus* at 4 dai.

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Table 3. The fungal growth on solid and liquid medium from seed

Period	Media	Diameter of Fungi (cm)						Average
		P. ostreatus		*G. virens*		*T. koningii*		
	Solid medium							
3 dai	Fresh extract champeden seed	4.45	a	7.33	cde	9,00	e	
	Powder champeden seed	4.03	a	6.5	bcd	8.33	e	
	Fresh extract durian seed	5.07	ab	7.63	de	5.72	abc	
	Powder durian seed	5.1	ab	5.32	ab	7.75	de	
	Fresh extract Indian almond seed	4.62	ab	8.00	de	7.97	de	
	Powder Indian almond seed	4.87	ab	7.42	cde	8.58	e	
	Fresh extract potato tuber	4.1	a	7.27	cde	9.00	e	
	PDA instant (kontrol)	4.67	ab	8.92	e	9.00	e	
	Average	4.61	a	7.3	b	8.17	b	
HSD 5%				l = 1.81				
4 dai	Fresh extract champeden seed	6.70		8.73		9.00		8.14
	Powder champeden seed	5.45		8.77		9.00		7.74
	Fresh extract durian seed	7.07		8.40		9.00		8.16
	Powder durian seed	5.93		7.82		9.00		7.58
	Fresh extract Indian almond seed	6.47		9.00		9.00		8.16
	Powder Indian almond seed	6.30		9.00		9.00		8.10
	Fresh extract potato tuber	5.38		9.00		9.00		7.79
	PDA instant (kontrol)	6.78		9.00		9.00		8.26
	Average	6.26	a	8.72	b	9,00	b	
HSD 5%				l = 0.50				
		Population of fungi (x 10^7 cfu/mL)						Average
	Liquid medium	*P. ostreatus*		*G. virens*		*T. koningii*		
4 wai	Fresh extract champeden seed	13,10	abcd	12,02	abcd	10,24	abc	11,79
	Powder champeden seed	7,19	ab	14,35	abcd	13,64	abcd	11,73
	Fresh extract durian seed	9,05	abc	7,06	ab	7,85	ab	7,99
	Powder durian seed	7,85	ab	21,20	de	16,79	abcd	15,28
	Fresh extract Indian almond seed	13,40	abcd	16,30	abcd	17,39	bcd	15,70
	Powder Indian almond seed	8,30	ab	29,24	ef	36,41	f	24,65
	Fresh extract potato tuber	8,81	abc	6,50	a	15,58	abcd	10,30
	PDA instant (kontrol)	8,81	abc	18,79	cd	14,46	abcd	14,02
	Average	9,56		15,68		16,55		

Note: Means followed by the same letter within a column are not significantly different according to Tukey's honestly significance difference (HSD), 5%

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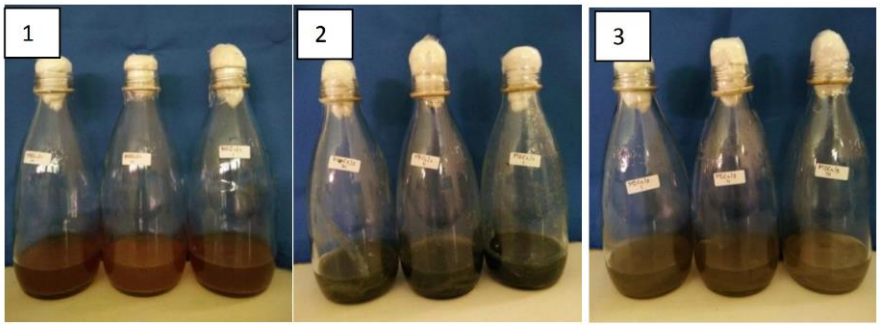

Figure. 3. Indian almond seed media (liquid media) is good media for growth (1). *P. ostreatus* (2). *G. virens*, and (3). *T. koningii*, at 4 wai.

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