Section A-Research paper
ISSN 2063-5346

Mycelial Biomass and Polysaccharide Production by *Ganoderma lucidum* (Fr.) P. Karst. (Fungi: Polyporaceae) Cultured on Mango Peel Extract

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ABSTRACT

Ganoderma lucidum is a highly medically important mushroom that has long been used as ingredient in health foods and drugs as it contains various bioactive compounds such as 1,3-β-D-glucan polysaccharides. Mango peels are wastes produced in large quantities in agricultural and industrial processes imposing disposal problems and environmental hazards. These mango peels were used as alternative cost-effective culture medium for mycelial submerged cultivation of *G. lucidum.* Three concentrations of mango peel extract were tested (10%, 50% and 100%). Shake-flask cultivation was used for culturing the fungus. After 7-day cultivation, produced mycelial biomass, extracellular polysaccharides (EPS) and intracellular polysaccharides (IPS) were quantified according to the methods used by Vukojević *et al.* (2006). There was a significant difference ($p < 0.05$) on the yields of mycelial biomass, EPS and IPS between the different puree concentrations of mango peel media and the controls, potato dextrose broth and water as positive and negative control media, respectively. The mean mycelial biomass yield in potato dextrose broth (1.733±0.715 mg/mL) was significantly higher compared to 50% (0.425 mg/mL) and 100% (0.342 mg/mL) MP puree and to water (0.258 mg/mL). However, the mean mycelial biomass yield in 10% and 100% MP extract, showed no significant difference which indicates that their yields were comparable. The EPS yield in 100% MP extract was significantly higher compared to each of the media. On the other hand, the IPS yield in 100% MP puree was significantly higher compared to the rest

of the media except to that of 50% MP puree. It was also observed that mycelial biomass was lower in higher puree concentration of mango peel media while both the EPS and IPS were higher at higher concentration of mango peel media. Hence, among the three puree concentrations of mango peel media, mycelial growth was maximal in 10%, while the production of EPS was most efficient in 100% and that of IPS was in 50% and 100%.

INTRODUCTION

Ganoderma lucidum, also known as "*Ling Zhi*" in China and "*Reishi*" in Japan, is a basidiomycete species belonging to family Polyporaceae, where members are distinguished by the presence of pores, instead of gills on the underside of the fruiting body (Erkel, 2009a). *G. lucidum* has received great interest in medicine because it is highly valued as ingredient in health foods, and most especially as component of drugs and treatment for various diseases. Moreover, it possesses numerous bioactive compounds such as polysaccharides and ganoderic acid that might be responsible for treating conditions like arteriosclerosis, diabetes, gastric ulcer, arthritis, cancer and many other diseases (Wagner *et al*., 2003; Vukojević *et al*., 2006). It also produces the polysaccharide 1,3-*β*-D-glucan that has high antitumor activity and inhibits a variety of cancers (Mizuno, 1999; Smith *et al*., 2002).

Several cultivation methods are established to obtain high yields and quality of *G. lucidum,* due to its medicinal significance, and its rareness in nature. Production of biologically active compounds can be obtained by mycelial cultivation after only 2-3 weeks without waiting for the full development of the fruiting body (Song *et al*., 2007; Wagner *et al*., 2003; Erkel, 2009b). In mycelial cultivation, nutritional components and environmental conditions (i.e., temperature and pH) are very important in maximizing the production of mycelial biomass and bioactive compounds (Wagner *et al*., 2003). Different substances and products such as whey permeate, starch, oil and ethanol have been tested and used as nutrient or additive for the culture media to enhance mycelial growth and bioactive compound production of the *G. lucidum* mycelia (Huang *et al*., 2008; Yang *et al*., 2004; Song *et al*., 2007).

Mango peel forms about 20% of the whole fruit and at present it is a waste product and its disposal has become a great problem, especially to the mango processing industries.

ISSN 2063-5346

The possibility of utilizing mango peels for the mycelial cultivation of *G. lucidum* is a sound solution to the problem of biowastes. Currently, different institutes such as the University of the Philippines Los Baños (UPLB) have conducted researches and projects exploring the use of mango peel so as to somehow minimize the environmental hazards it causes. However, its use in the biotechnological production of
Ganoderma lucidum has not yet been explored. Thus this study aims to use mango peel extracts as medium in shake-flask cultivation of *Ganoderma lucidum* to produce mycelial biomass and polysaccharides. Furthermore, determine the optimum concentration of mango peel puree in media that maximizes growth and production of intracellular and extracellular polysaccharides of *Ganoderma lucidum*.

MATERIALS AND METHODS

Microorganism and Culture Media

Mycelia of *Ganoderma lucidum*, were acquired from the Plant Protection Section (PPS), Visayas State University (VSU), Baybay, Leyte, Philippines, maintained on malt extract agar (MEA) in petri dishes at 30°C. Sub-culturing was done once every month and then stored at 4°C (Wagner *et al.,* 2003). Potato Dextrose Broth (PDB) was used for the pre-culture of mycelial suspension. In the subsequent cultivation three different mango peel puree (extract) concentrations of 10%, 50% and 100% of mango peel media, including potato dextrose broth (PDB), as a positive control, and distilled water, as negative control were used. Fresh ripe mango peels were obtained from Profood International Corporation, Maguikay, Mandaue City, Cebu, Philippines. The peels were washed with sterile distilled water. They were chopped or cut into small pieces by using knives. One hundred grams of mango peelings and 200 ml of distilled water were mixed and grinded using a blender. Solid particles were separated from the extract through filtration using cheesecloth. These concentrations of mango peel puree were chosen so as to represent low, medium and high concentrations, respectively. Two hundred ml of each aliquot and control media were placed in 500 ml Erlenmeyer flasks, no other nutrients were added. The initial pH of the media were adjusted to 6.5 (0.05M NaOH and 0.05M HCl) using an Orion 3 star pH meter (sensitivity: + 0.01 pH units) (Fang and Zhong, 2002a). The flasks

were then plugged with cotton and covered with aluminum foil and were sterilized in an autoclave at 121°C, 15 pounds per square inch for 15-20 minutes.

Shake-flask Cultivation

G. lucidum was pre-cultured, three mycelial blocks (approximately 10 mm in diameter) from the stock culture inoculated on a 200 ml PDB in 500 ml Erlenmeyer flask using a sterile inoculating needle. The cultures were plugged with cotton and incubated at room temperature and shake at 150 rpm using an Edmund Buhler rotary shaker for 7 days without illumination (Nasreen, *et al.,* 2005; Wagner, *et al.,* 2003; Yang and Liau, 1998; Fang and Zhong, 2002a). Twenty ml (i.e. 10% of the total volume of the medium in the flask) of the soft and spiny mycelial pellets from the pre-culture was inoculated to each flask containing 200 ml of the different aliquots and controls. The cultures with 6 replicates were plugged with cotton and incubated at room temperature and shake at 150 rpm using an Edmund Buhler rotary shaker for 7 days without illumination (Nasreen, *et al.,* 2005; Wagner, *et al.,* 2003; Yang and Liau, 1998; Fang and Zhong, 2002a).

Analysis of Culture Parameters

Mycelial biomass. To obtain the mycelial biomass, cultures were filtered using a vacuum filter, washed off the filter with deionized water, centrifuged at 3000 rpm for 30 minutes at room temperature and oven-dried at 100°C until the weight is constant. The final weight was determined and presented as mg.mL^{-1} of medium (Vukojević *et al.*, 2006).

Extracellular polysaccharides (EPS). Extracellular polysaccharides are high-molecular-weight polymers that are composed of sugar residues and are secreted by an organism into the surrounding environment. Since mango peels also contain polysaccharides, the EPS of the mango peel media was quantified prior to the 7-day cultivation period and was deducted from the amount of EPS quantified from that of the mango peel media with inocula after the cultivation period to get the actual EPS produced by *G. lucidum.* To quantify EPS, twenty ml was withdrawn from each of the culture and centrifuged at 13000 rpm for 30 minutes at room temperature. The supernatant was dialyzed and precipitated by adding four volumes of 95% ethanol at 4°C. Then, it was centrifuged again (3000 rpm, 10 min, 4°C) to separate the precipitate. The produced EPS were oven-

dried at 50°C to constant weight and were presented as mg.mL^{-1} of supernatant (Vukojević *et al.*, 2006).

Intracellular polysaccharides (IPS). Intracellular polysaccharides are polymers composed of sugar residues found within the mycelial cells. To quantify IPS, twenty ml, withdrawn from the homogenized culture, was centrifuged at 13000 rpm for 30 minutes at room temperature. The collected mycelial pellets were then weighed, dried, frozen, and macerated. Produced intracellular polysaccharides (IPS) were extracted from the collected pellets by cooking in 10 ml of distilled water at 100°C for one hour. After cooling, the supernatant was separated by centrifugation (4°C, 3000 rpm, 30 min), dialyzed and precipitated by adding four volumes of 95% ethanol at 4°C. Then, it was centrifuged (3000 rpm, 10 min, 4°C) again. The amount of produced IPS was presented as mg.g^{-1} of dried mycelial weight (Vukojević *et al.*, 2006).

Statistical Analysis

One-way ANOVA was used to determine and verify whether the yields of mycelial biomass, intracellular and extracellular polysaccharides were significantly different among the various culture media (mango peel media with different puree concentrations and the controls). The significance level was set at $\alpha = 0.05$. Post hoc Tukey HSD test followed. Results of the statistical analysis were presented in a statistical table. Mean values of the quantified mycelial biomass, IPS and EPS in different media were presented in a bar graph.

RESULTS

Mycelial Growth

It was observed that *Ganoderma lucidum* grew on the mango peel media, as well as in the water (- control). The mycelial growth was observed to be in spherical form with spines and in varying sizes. The mycelia were in whitish or yellowish to brownish in color (Figs. 1a and b).

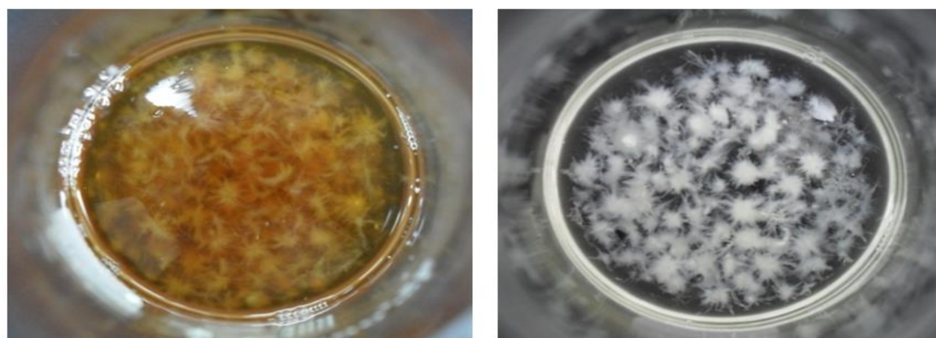

Figure 1. Mycelial growth of *G. lucidum* in (**a**) mango peel media (10% puree concentration) and (**b**) water (top view of the flask).

Mycelial Biomass

Mean mycelial biomass of *Ganoderma lucidum* on different culture media is shown in Figure 2. Maximum mycelial growth (1.733 mg/mL) was recorded on potato dextrose broth (PDB). However, among the mango peel media with various concentrations of mango peel puree, highest biomass density (0.717 mg/mL) was obtained at 10% while the least biomass density (0.342 mg/mL) was at 100%. The biomass density (0.258 mg/mL) obtained from the water media (negative control) was quite close to that of 100% mango peel puree.

Mean mycelial biomass yield differed significantly between the different culture media (One-way ANOVA, $P<0.05$; Table 1). Particularly, the mean biomass density in PDB (+ control) was significantly higher compared to that of the other media except for the media containing 10% mango peel puree (Tukey HSD Test, $P=0.8017$; Table 2).

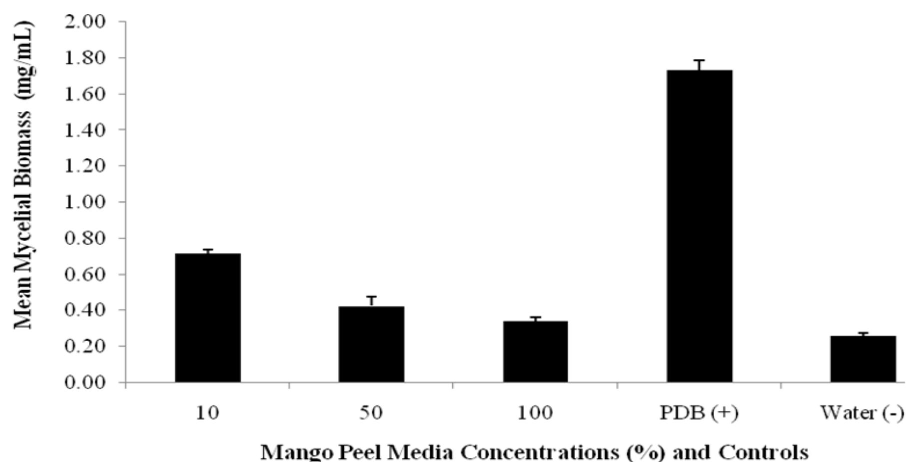

Figure 2. Mean mycelial biomass (+SD) of *Ganoderma lucidum* on different media.

Table 1. One-way ANOVA of the mean mycelial biomass of *Ganoderma lucidum*

Source of Variation	*df*	*SS*	*MS*	*F*	*P-value*	*F crit*
Between Groups	4	4.4483	1.1121	5.3904	0.0012	2.5787
Within Groups	45	9.2837	0.2063			
Total	49	13.7321				

Table 2. Tukey HSD Test for Differences between means of the mycelial biomass yield of *G. lucidum*

Groups	*Difference*	*Test Statistics*	*P-value*
10 vs 50	0.175	1.2184	0.9094
10 vs 100	0.225	1.5665	0.8017
10 vs PDB	-0.54	3.7596	0.0767
10 vs Water	0.275	1.9146	0.6597
50 vs 100	0.05	0.3481	0.9992
50 vs PDB	-0.715	4.978	0.0085*
50 vs Water	0.1	0.6962	0.9877
100 vs PDB	-0.765	5.3261	0.0042*
100 vs Water	0.05	0.3481	0.9992
PDB vs Water	0.815	5.6742	0.0021*

*significantly different between the two media

Extracellular Polysaccharides

Figure 3 shows the mean extracellular polysaccharide (EPS) yield of *G. lucidum* on different culture media. Highest EPS yield (4.156 mg/mL) was obtained in the mango peel media with 100% mango peel puree concentration. It was decreasing as the puree concentration of the mango peel media decreases. Thus, among the three puree concentrations, the lowest EPS yield (0.178 mg/mL) was obtained at 10% while the highest yield (4.156 mg/mL) was obtained at 100%. In PDB, the EPS yield (1.233 mg/mL) was relatively lower than that in media containing 50% and 100% concentration

of mango peel puree. In water, however, the EPS yield (0.689 mg/mL) was relatively higher compared to 10% mango peel media.

Mean extracellular polysaccharide yield showed a significant difference between different culture media (*P*<0.05; Table 3). The EPS yield in the media with 10% mango peel puree was significantly higher compared to that of 10%, 50%, PDB and water (*P*=0.0003, *P*=0.0343, *P*=0.0093, *P*=0.0015, respectively; Table 4).

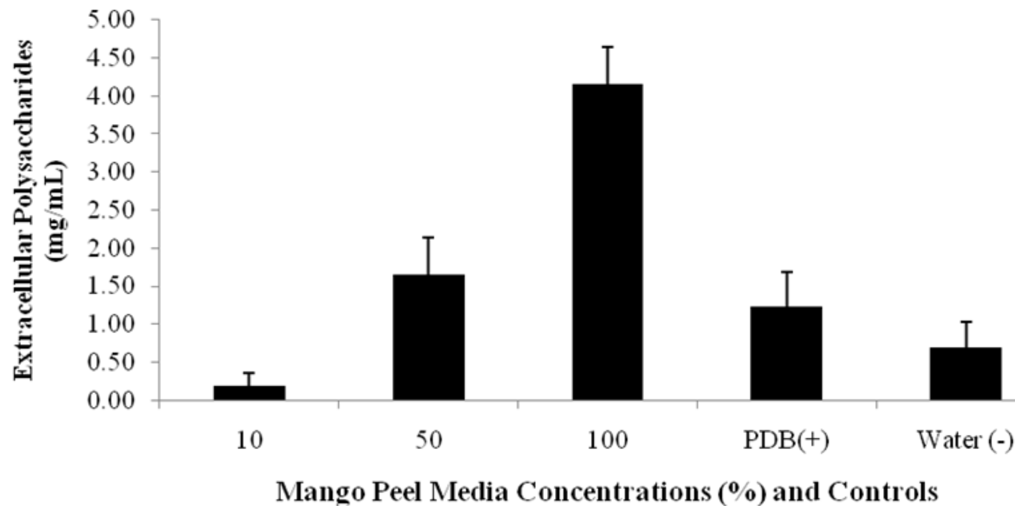

Figure 3. Mean extracellular polysaccharide yield (+SD) of *Ganoderma lucidum* on different culture media.

Table 3. One-way ANOVA of the mean extracellular polysaccharide yield of *G. lucidum*

Source of Variation	*df*	*SS*	*MS*	*F*	*P-value*	*F crit*
Between Groups	4	34.2629	8.5657	6.7702	0.0002	2.5787
Within Groups	45	56.9343	1.2652			
Total	49	91.1972				

ISSN 2063-5346

Table 4. Tukey HSD Test for Differences between means of the extracellular polysaccharide yield of *G. lucidum*

Groups	*Difference*	*Test Statistics*	*P-value*
10 vs 50	-0.8799	2.4737	0.4151
10 vs 100	-2.3866	6.7096	0.0003*
10 vs PDB	-0.6333	1.7804	0.7172
10 vs Water	-0.3067	0.8622	0.9729
50 vs 100	-1.5067	4.2359	0.0343*
50 vs PDB	0.2466	0.6933	0.9879
50 vs Water	0.5732	1.6115	0.7849
100 vs PDB	1.7533	4.9292	0.0093*
100 vs Water	2.0799	5.8474	0.0015*
PDB vs Water	0.3266	0.9182	0.9659

*significantly different between the two media

Intracellular Polysaccharides

The mean intracellular polysaccharide yield of *G. lucidum* is shown in Figure 4. The same trend was observed to that of EPS yield. It was decreasing with decreasing concentration. Hence, among the three concentrations of mango peel media, IPS yield (554.824 mg/g of dried mycelial weight) was maximal in media with 100% mango peel puree concentration while it was lowest in 10% concentration (65.055 mg/g). However, the IPS yield in PDB (83.769 mg/g) was just slightly higher than that of 10% conc. The lowest yield (46.14 mg/g) among all the media was observed on the negative control – water.

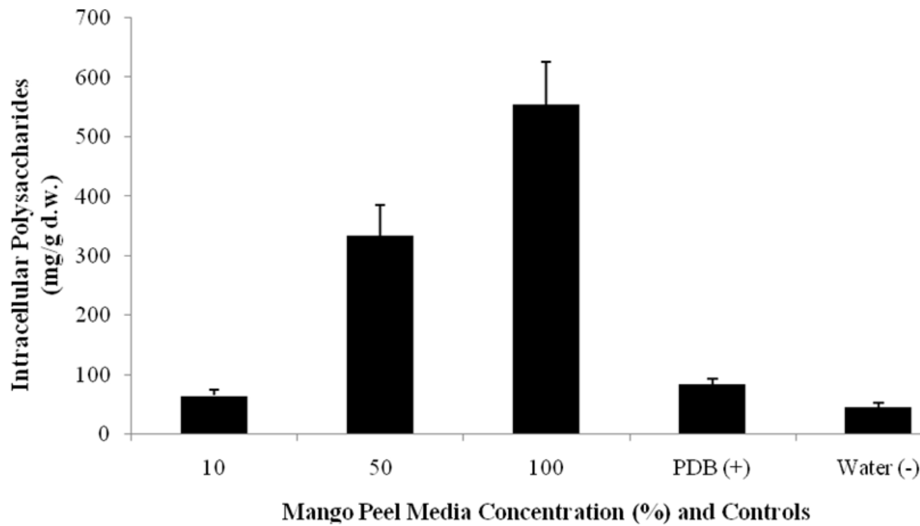

Figure 4. Mean intracellular polysaccharide yield (+SD) of *Ganoderma lucidum* on different culture media.

There was a significant difference on the intracellular polysaccharide yield of *G. lucidum* between different culture media ($P<0.05$; Table 5). The IPS yield in 100% mango peel media was significantly higher compared to that of 10%, PDB and water (P=0.0011, P=0.0017, P=0.0007, respectively; Table 6). Whereas there was no significant difference to the IPS yield between 50% and 100% (P=0.3272; Table 6).

Table 5. One-way ANOVA of the mean intracellular polysaccharide yield of *G. lucidum*

Source of Variation	*df*	*SS*	*MS*	*F*	*P-value*	*F crit*
Between Groups	4	712,585.79	178,146	7.4287	0.0001	2.5787
Within Groups	45	1,079,137	23,980.82			
Total	49	1,791,723				

Table 6. Tukey HSD Test for Differences between means of the intracellular polysaccharide yield of *G. lucidum*

Groups	*Difference*	*Test Statistics*	*P-value*
10 vs 50	-161.61	3.3002	0.1533

10 vs 100	-293.86	6.0008	0.0011*
10 vs PDB	-11.229	0.2293	0.9999
10 vs Water	11.3474	0.2317	0.9998
50 vs 100	-132.25	2.7006	0.3272
50 vs PDB	150.385	3.0709	0.2092
50 vs Water	172.961	3.532	0.1093
100 vs PDB	282.633	5.7715	0.0017*
100 vs Water	305.209	6.2325	0.0007*
PDB vs Water	22.5759	0.461	0.9975

*significantly different between the two media

DISCUSSION

Ganoderma lucidum's mycelial growth exhibited a pelleted growth form (Figs. 1a and b). Pellet formation in submerged culture is generally recognized to be greatly influenced by the type of inocula, composition of the culture medium and cultural conditions. The most important advantage of growth in the pellet form is attributed to a considerable decrease in the viscosity of medium, in comparison to filamentous form. However, cultivation in pellet form may, in some cases, introduce undesirable limitations and the most important of these is the diffusional limitations of essential nutrients like carbohydrates and also oxygen into the deeper layers of the mycelial pellets (Scrinivasan, 2004).

Mycelial growth was unexpectedly evident in the negative control media – water – which was quite comparable to the growth observed in the mango peel media with 100% puree concentration (Figs. 1b and 2). The type of inoculum used may account for this result. Since the inoculum was taken from a liquid culture medium, specifically, potato dextrose broth (PDB), little amount of the medium, which contains nutrients, was transferred to the water media allowing slight yet discernible mycelial growth in water.

The results on the mycelial biomass yield indicate that *Ganoderma lucidum* can grow in a media made of mango peel. Among the three concentrations of mango peel puree on media, it was in 10% that the biomass density was highest (Fig. 2). In addition, there was a decrease in the average biomass density of *G. lucidum* as the mango peel puree concentration increases. These results can be attributed to the effect of initial sugar

concentration in the medium. Mango peel contains a lot of carbohydrates (Ajila *et al.,* 2006). Readily utilizable sugars such as glucose frequently exert 'carbon catabolite' inhibition of product formation of fungi (El-Mancy and Bryce, 1999). Additionally, Fang and Zhong (2002b) obtained the same trend in their study. They attributed the less efficiency of the growth of *G. lucidum* at higher sugar concentration to the increased osmotic pressure in the medium that slows down fungal growth.

The lack of significant difference between the mycelial biomass obtained from the PDB (positive control media) and in the media with 10% mango peel puree (Table 2) signifies that the growth in both media are comparable.

The results showed that both extracellular and intracellular polysaccharide yield increased as the mango peel puree in media increases (Figs. 3 and 4). Several factors that may vary between the media contribute to the product formation including initial sugar concentration and aeration. Relative productivity of the various metabolites depends on the pellet size, which in turn is reliant on the initial sugar concentration of the media. Polysaccharide concentrations, for example, are lower in larger pellets and higher at a lower dissolved oxygen tension (DOT; Wagner *et al*., 2003). These factors, however, were not controlled nor monitored in this study.

When comparing the mycelial biomass density to the production of polysaccharides in the different mango peel puree concentrations in media, it was decreasing as the concentration increases in contrast with that of polysaccharide production, which was increasing with increasing concentration (Figs. 2, 3 and 4). The dissimilarity in the rate of nutrient assimilation of *G. lucidum* in varying concentration of media accounts for this difference. Where the concentration is high, nutrient assimilation is low since the nutrients are not scarce. This results to slow mycelial growth yet high production of metabolites. On the other hand, in low concentrated media where nutrients are insufficient, the organism tends to assimilate the nutrients faster; thereby resulting to fast mycelial growth yet low metabolite production. Thus, when the organism devotes its energy to reproduction and growth, metabolite production is low; conversely, when the organism is allocating its energy more on metabolite production, growth is minimized.

In the study of Vukojević *et al.* 2006 (Table 7), the mycelial biomass yield was a great deal higher than that of our study. The EPS yield, on the other hand, was comparable, as

well as that of IPS except in the potato dextrose broth and 10% mango peel media used in our study, where the quantified amounts were relatively higher by more than twice. However, generally, their results conform to ours in that in media with high nutrient (i.e. carbon) concentration, mycelial biomass was high while both EPS and IPS yields were low. Nutrient assimilation of *G. lucidum* in media with low nutrient concentration is fast due to the limited nutrient supply which therefore results to faster mycelial growth. Under this condition, the organism tends to allocate its accumulated nutrients in reproduction rather than in the production of metabolites.

Nasreen *et al.* (2005) obtained higher yields of mycelial biomass particularly in potato dextrose broth (15.90 mg/mL) compared to our study using the same media (1.73 mg/mL; Table 7). These results suggest that cultivation period is significant for accumulation of mycelial biomass (Vukojević *et al.,* 2006). Nasreen *et al.* (2005) used longer cultivation period (15 days) than in our study (7 days). If the mycelia of *G. lucidum* in our study were given a longer culture time, produced biomass density may be comparable to that obtained by Nasreen *et al.* (2005).

Table 7. Liquid-state cultivation undertaken with *Ganoderma lucidum* and key results obtained

Media	Working volume and conditions	Maximum concentrations o			Author
		MB	EPS	IPS	
		mg/mL	mg/mL	mg/g	
Synthetic medium (composition*)	50 mL in 250-mL flasks; pH 5.0 (MB), pH 3.0 (EPS), pH 5.5 (IPS); 160 rpm on rotary shaker; room temperature; inoculum 10%	20.60	5.20	69.93	Vukojević *et al.* (2006)
Kirk[8] Media (A) + molasses	100 mL in 250-mL flasks; pH 5.0; 100 rpm on rotary shaker; 25°C; disk inoculum	6.80	-	-	Nasreen *et al.* (2005)
Kirk[8] Media (A) + Glc		3.80	-	-	
PDB		15.90	-	-	
ME		9.10	-	-	
B + Glc; YE; P	50 mL in 250-mL flasks; initial pH	16.70	1.08	-	Fang and

	5.5; rotary shaker 120 rpm; 30°C. Inoculum 10%				Zhong (2002b)
B + lactose; YE; P	3.5 L in bioreactor; 30°C; 200 rpm; inoculum*	15.60	0.92	-	Tang and Zhong (2003)
C + safflower oil	100 mL in 250-mL flask; initial pH=4; 100 rpm on rotary shaker; 30°C. Disk inoculum	2.78	0.18	-	Yang *et al.* (2000)
C + oleic acid		4.60	0.16		
PDB	200mL in 500-mL flasks; pH 6.5; 150 rpm on rotary shaker; room temperature; inoculum 10%	1.73	1.23	83.77	Cercado *et al. (2010)*
MPM 10%		0.72	0.18	65.05	
MPM 50%		0.43	1.64	334.41	
MPM 100%		0.34	4.16	554.82	

*not possible to determine from the article

A = KH_2PO_4; $MgSO_4$; $CaCl_2.2H_2O$; $(NH_4)_2SO_4$; YE

B = $KH_2PO_4.H_2O$; $MgSO_4.7H_2O$; vitamin B1

C = Glc; K_3HPO_4; KH_2PO_4; $MgSO_4.7H_2O$; YE; NH_4Cl

Abbreviations: MB = mycelial biomass; EPS = extracellular polysaccharide; IPS = intracellular polysaccharide; Glc = glucose;

PDB = potato dextrose broth; ME = malt extract; YE = yeast extract; P = peptone; MPM = mango peel media

In the study of Fang and Zhong (2002b) and Tang and Zhong (2003), higher mycelial biomass (16.7 mg/mL and 15.6 mg/mL respectively) were also obtained in comparison to our study (Table 7). These results suggest that nutrient content of the media greatly influences mycelial growth. The growth of the *G. lucidum* mycelia is enhanced when the media are added with complex carbon and nitrogen source such as yeast extract, peptone or malt extract (Wagner *et al.*, 2003). This was evident in the studies of Fang and Zhong (2002b) and Tang and Zhong (2003), in which yeast extract and peptone were added in both studies while different carbon sources, glucose for the former while lactose for the latter, were added. However, higher concentrations of these nutrient sources inhibit cell growth.

Yang *et al.* (2000) obtained slightly higher mycelial biomass and EPS yield compared to our study (Table 7). This suggests that fatty acids have effect on the product formation of *G. lucidum* (Wagner *et al.,* 2003; Yang *et al.,* 2000). The media used in their study was synthetic added with safflower oil and oleic acid. Two types of safflower oil exist, one that is rich in oleic acid and one that is rich in linoleic acid. Oleic acid resulted in high biomass density. Linoleic acid had a strong inhibitory effect, however Yang *et al.* (2000) pointed out that in their study, linoleic acid stimulated both growth and EPS production, which was evident in the results they obtained. They implied that other components of the oil might be acting positively, masking the inhibitory effect of linoleic acid.

In terms of mycelial biomass yield, generally, the results in this study was relatively lower compared to that obtained in the studies of Vukojević *et al.* (2006), Nasreen *et al.* (2005), Fang and Zhong (2002b), Tang and Zhong (2003) and Yang *et al.* (2000). In all these previous studies, synthetic media were used in cultivating the mycelia of *G. lucidum* unlike in this present study where organic medium was used. Synthetic media are formulated in a way that the nutrients present are in readily utilizable forms (i.e. monosaccharides), which can be easily absorbed by the organism. When the nutrient assimilation of the organism is fast, its growth will be rapid. Hence, there were higher yields of mycelial biomass obtained in the said studies.

CONCLUSIONS

The mycelia of *Ganoderma lucidum* were grown on mango peel media of different puree concentrations (10%, 50% and 100%). The production of mycelial biomass, extracellular polysaccharides and intracellular polysaccharides by *G. lucidum* was quantified after 7-day shake-flask cultivation. The quantification method was based on Vukojević *et al.*'s (2006).The production rate of the desired products among different culture media was compared and the presence or absence of a difference between them was determined. With the results obtained, it was concluded that the mycelia of *G. lucidum* can grow on the mango peel media. Mycelial biomass yield was higher in lower concentrations of mango peel media. On the other hand, extracellular polysaccharide yield was higher in higher concentrations of mango peel media. The same trend was observed in the yield of intracellular polysaccharides.

Mango peels, therefore, can be used as a cost-effective media for the production of mycelial biomass, extracellular polysaccharides and intracellular polysaccharides of *Ganoderma lucidum.*

For future pursuance of this study, the following are recommended for the improvement of this research. Determination of the nutrients present in the mango peels should be included so as to know the specific nutrients responsible for the enhanced mycelial growth of *G. lucidum* and those that inhibit its growth. A wider range of the concentrations of the mango peel media should be tested. Bioreactor tanks should be used for the cultivation of the organism for easier control of the conditions (i.e. pH and temperature) affecting the growth. Quantification of ganoderic acid should also be included. Daily monitoring of the nutrient (i.e. carbohydrate) content of the media should be done to know how much nutrient is depleted per day. Regular monitoring of the desired products (i.e. polysaccharides and ganoderic acid) should be done so as to know the trend of the production rate throughout the cultivation period. Cultivation period should be longer (2-3 weeks).

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