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Mycelial Growth of *Ganoderma curtissii* in Locally Indigenous Media

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Abstract

Fungal study requires culture medium for evaluation of its mycelial form and storing viable cell lines. In this study innovative media such as coconut water from matured nuts (CW), corn grit (CG) and rice bran (RB) decoction was evaluated. The ideal media for luxuriant growth of Ganoderma curtissii was coconut water media at physical condition of 26.40 ^{O}C and 81.29% relative humidity respectively.

Keywords: *Ganoderma curtissii*, coconut water, corn grit, rice bran, mycelial form, relative humidity

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1. Introduction

Media is a critical aspect in growing mushrooms. It is a way of storing viable cell lines of microorganism, wherein this cell lines can lead to various breakthrough for the benefits of mankind. For instance, the genus *Ganoderma* species of mushroom contains bioactive components with antioxidant properties [1]. Since early 2,000 years ago it has been used in traditional Chinese herbal medicine which is then extended worldwide [2]. Reference [3] claimed that pharmacological properties of *Ganoderma* have been associated with its ability to reduce the risk of heart disease, cancer and stimulate the immune system,

Furthermore, [4] added that its health beneficial properties are attributed to the bioactive components such as polysaccharides, triterpenes, sterols, lectins and some protein. Reference [1] stated that recently the use of fungi of the genus *Ganoderma* has become increasingly important in the human diet for its nutritional and pharmacological characteristics. These breakthroughs generally begin via rescuing of healthy cell lines of fungi which was being cultured in medium. Though fungi inhabit every possible environment [5] it requires medium which will provides its nutrient requirement. However, all microbiological media cost is rising fast [6]. Generally, fungal cultures are

grown on potato dextrose agar (PDA), Sabouraud dextrose agar (SDA), or cornmeal agar (CMA) which is very expensive. Thus, evaluation on potential locally available and abundant resources is needed to further researches under this field.

2. Objectives of the Study

This study aimed to evaluate the luxuriant mycelial growth performance of *G. curtissii* in various potential locally available materials for *G. curtissii* culture media.

3. Materials and Methods

3.1. Source of *G. curtissii*

The pure culture of *G. curtissii* was obtained at Center for Tropical Mushroom Research and Development, College of Arts and Sciences, Central Luzon State University, Science City of Munoz Nueva Ecija, Philippines.

3.2. Mycelial growth of *G. curtissii* in various medium

Mycelial growth of *G.curtissii* was evaluated in three indigenous media; (CW) Coconut water from matured nuts, (CG) corn grit and (RB) rice bran. Wherein 50 grams of corn grit and rice bran were added with one liter of tap water and boiled for 15 minutes. Subsequently, the decoction was collected, it was volume up to attain one liter of solution and added with 10 grams of white table sugar and 20 grams of white gulaman bar. It was boiled until homogenous mixtures were attained; subsequently it was dispensed in a 1500 ml Erlenmeyer flask and secured with cotton plug. Meanwhile the coconut water was added with 20 grams of white gulaman and boiled until homogenous mixture was attained. It was dispensed in a 1500 ml Erlenmeyer flask and sealed with cotton plug. The prepared media were sterilized at 121 °C for 20 minutes.

3.3. Pour plating and inoculation

The newly sterile media was dispensed in a petri plates and allowed to cool. Then, seven day old ten mm mycelial block was aseptically inoculated on the center of the media and allowed to ramify. The data were gathered via daily measurement of the mycelial run until the media is fully colonized.

3.4. Statistical analysis

The statistical design used was completely randomized design using Statistical Tool for Agricultural Research (STAR) model.

4. Results and Discussion

4.1. Daily mycelial performance of *G. curtissii* in different culture media

Mycelia are the masses of hyphae that are threadlike in structure which is considered as the vegetative part of a fungus [7]. Luxuriant production of mycelial run of *G. curtissii* was evaluated in this study with the emphasis on readily locally available innovative media. Mycelial performance in coconut water (matured nuts), corn grit and rice Bran media were evaluated. Results shows that 6th and 7th day of mycelial run on CG reveals no significant difference ($P < 0.05$); however, from day one to five significant differences on the mycelial ramification of *G. curtissii* were recorded. Meanwhile CW mycelial growth in 6th day was significantly different ($P < 0.05$) among the number of days of mycelial ramification except from 5th day. Moreover, in terms of RB statistical analysis reveals no significant differences ($P < 0.05$) with regards to number of days of mycelial colonization were noted.

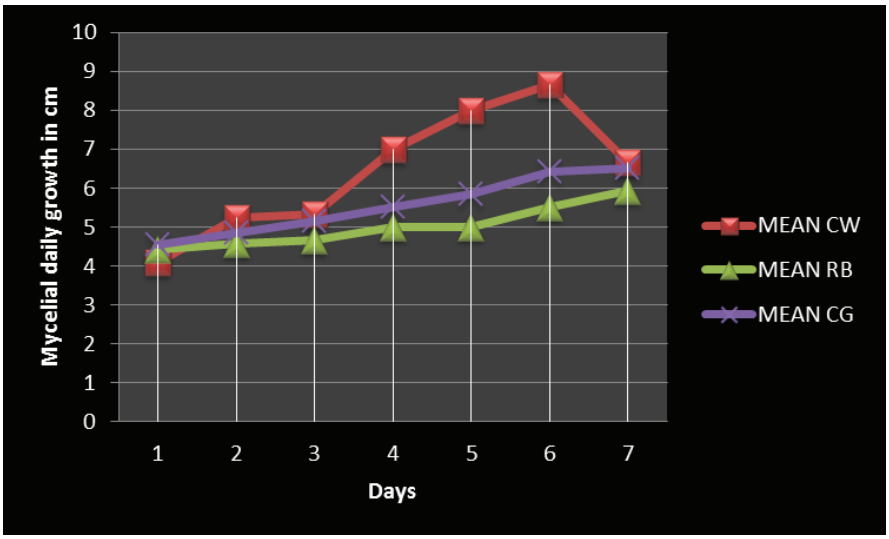

Figure 1: Daily mycelial growth of *G. curtissii*.

4.2. Mycelial colonization of *G. curtissii* in culture media

Media is vital in mycelial production of mushrooms, since it serves as an agent to store viable cell lines of fungi. No significant (P < 0.05) difference were noted in mycelial run of *G. curtissii* in CG, CW and RB from the first day to third, however, significant result were noted on the fourth and fifth day of colonization. Furthermore, there was no significant (P < 0.05) difference noted on the seventh day of mycelial colonization in CG and CW but not in RB. The consistent growth of CW can be attributed to its nutrients components which is 4.41% carbohydrates, 0.52% protein, and 0.15% fat [8].

TABLE 1: Mycelial Growth of *G. curtissii* in Culture Media.

Culture Media	Number of Days						
	1	2	3	4	5	6	7
CG	4.5833a	4.8333a	5.1667a	5.5000b	5.8333b	6.4167b	6.5000a
CW	4.0833a	5.2500a	5.3333a	7.0000a	8.0000a	8.6667a	6.6667a
RB	4.0833a	4.2500a	4.8333a	5.0833b	5.0000b	5.1667c	5.3333b

* Means with the same letter are not significantly different

4.3. Number of days of *G. curtissii* full ramification

Period of days of full media colonization is essential in storing significant cell lines of microorganism since the shorter the period to an organism required to fully colonized a media equate to the lesser the chances of undergoing possible contamination of pure cultures. In terms of total number of days of full mycelial colonization of *G. curtissii* no sigificant difference (P < 0.05) were recorded interms of CG, CW and RB. However, CW shows the shortest number of days to fully colonized the media with 7.33 days, CG reveals 8.33 days while the longest number of days were recorded in RB with 9.67 days. This can be atributed 0.29% protein, 22.0 % carbohydrates [9] and cytokinin [8] that coconut water contains. Furthermore, the thickest mycelia were noted in RB however it took 9.67 days to fully colonize the medium. However coconut water elucidated the fastest mycelial run with visible zonation pattern in the medium (Figure 2). The highest mycelial growth of CW can be attributed to its chemical composition wherein according to Snowdon et al. [10], water contains protein (0.3 g), potassium (310 mg) and iron (1.1 mg) while [11] noted the presence of nitrogen (0.05%) and calcium oxide (0.69%); in addition, it contains Vitamins, minerals, amino acids, enzymes, growth factors, and trace elements such as zinc, selenium, iodine, sulfur, manganese, boronerl molybdenum [12]. This result is in congruent with the findings of [13] wherein coconut water is an ideal medium for Lintinus tigrinus, Schizophyllum commune [14] and *Pleurotus*

djamor [7]. Moreover, the poor ramification of RB can be attributed to the physical factor gathered which are 26.4° C and 81.29% RH since the favorable medium temp for mycelial growth of *Ganoderma* is 30°C [15] and an RH of 90 - 95% [16].

Table 2: Total Mycelia Colonization of *G. curtissii* in Media.

TRT	TNDMR	MD
CW	7.33 a	+++
RB	9.67 a	++++
CG	8.33 a	++

*TNDMR = total number of days of mycelial ramification
MD = mycelial density : (+) very thin, (++) thin,
(+++) thick, (++++) very thick

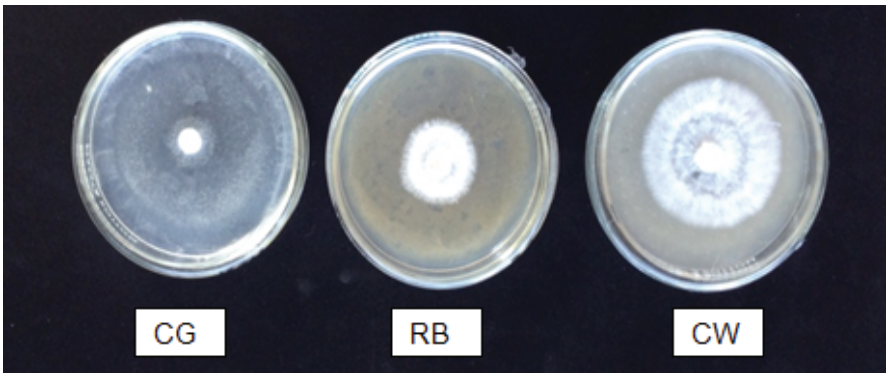

Figure 2: *G. curtissii* mycelial run at 5 days of incubation.

5. Conclusion and Recommendation

Findings of the study confirmed that coconut water from matured nut, rice bran and corngrit decoction can sustain mycelial growth of *G. curtissii*. However, the ideal media was coconut water as compared to the test media in terms of mycelial density and ramification of *G. curtissii* at 26.4°C and 81.29% RH physical condition.

Coconut water from matured coconut is highly recommended for isolation media for *G. curtissii* since it is locally available and abundant in the Philippines and because it is high on protein, carbohydrates (Ullah et al., 2010) and cytokinin (Yong et al., 2009).Different concentration of rice bran and corn grit as source of decoction is highly recommended for further studies.

References

[1] Huerta A. I., Molina T. J., Garnica R. Ma. G. and Yahuaca J.B. (2016). Total Polyphenols and Antioxidant Activity of Ganoderma curtissii extracts. Journal of Medicinal Plants Studies 4(4): 136 – 141.

[2] Wachtel-Galor S, Buswell J. A, Tomlinson B, Benzie I. F. F. Lingzhi.(2004). polyphorous fungus. In: Herbal and Traditional Medicine: Molecular Aspects of Health. New York: Marcel Dekker Inc. pp. 179-228.

[3] Russell R, and Paterson M. Ganoderma, a therapeutic fungal biofactory. Phytochemistry. (2006) 67:1985-2001.

[4] Ferreira ICFR, Vaz JA, Vasconcelos MH, and Martins A. (2010). Compounds from wild mushrooms with antitumor potential. Anti-cancer Agents in Medicinal Chemistry. 10:424-436.

[5] Nasim G, Malik S. M., Bajwa., Afzal, M and Mian, W. (2001). Effect of Three Culture Media on Mycelial Growth of Oyster and Chinese Mushroom. Asian Network for Scientific Information. Online Journal of Biological Sciences 1 (2): 1133.

[6] Arushdeep S. and Farooq U. (2014). Sugarcane Bagasse: A Potential Medium for Fungal Cultures. Chinese Journal. Volume 2014. Article ID 840505.

[7] Zurbano L. Y., Bellere A. D. Savilla L.C. (2017). Mycelial Growth, Fruiting Body Production and Proximate Composition of Pleurotus djamor on Different Substrate. The CLSU International Journal of Science and Technology. Volume 2.No. 1: 7-6. DOI: 10.22137/ijst.2017.v2nl.03.

[8] Yong J.W.H., G.e. L. Ng. and S.N. Tan. (2009). The Chemical Composition and Biological Properties of Coconut (Cocos nucifera L.) Water. 14, 5144-5164; doi:10.3390/molecules14125144.

[9] Ullah, I., M. Ali and A. Farooqi. (2010). Chemical and nutritional properties of some maize (Zea mays L.) varieties grown in NWFP, Pakistan. Pakistan Journal of Nutrition. 9(11): 1113-1117.

[10] Snowdown, W., T. Osborn, B. Aarberslsberg. and J. Schultz. (2003). Coconut and its role in health. Secretariat of the Pacific Community. Cataloguing-in-publication data. 23pp.

[11] Campbell, D., T. Thomas, T.M. Falck, N. Tutuo and K. Clem. (2000). The intravenous use of coconut water. American Journal of EmergencyMedicine 18:108–111.

[12] Fife B. (2008). Nutritional benefits of coconut water. Retrieved from the World Wide Web: http://www.litalee.com/ shopexd.asp?id = 388.

[13] Dulay R.M.R., S. P. Kalaw, R.G.R. Reyes, E. C. Cabrera, and N.F. Alfonso. (2012). Optimization of Culture Conditions for Mycelial Growth and Basidiocarp Production of Lentinus tigrinus (Bull.) Fr., A new Record of Domesticated Wild Edible Mushroom in the Philippines. Philippine Agric Scientist, Vol. 95 No. 3, 278 – 285, ISSN 0031 – 7454.

[14] Reyes R, Grassel W. and Rau U. (2009). Coconut water as a novel culture medium for the biotechnological production of schizophyllan. J Nature Stud 7(2):43–48.

[15] Jayasinghe C., Iimtiaj A., Hu H., Lee G.W., Lee T. S. and Lee U.Y. (2008). Favorable Culture Conditions for Mycelial Growth of Korean Wid Strains in Ganoderma Lucidum. Mycobiology 36(1): 28-33. PMCID: PM3755248.

[16] Lisieka J., Rogalski J., Sobieralski K., Siwulski M., Sokol S. and Ohga S. (2015). Mycelium Growth and Biological Efficiency of Ganoderma lucidum on Substrate Supplemented with Different Organic Additives. J.Fac. Agr., Kyushu Univ. 60 (2), 303 – 308.