COMPERATIVE STUDY ON THE PROXIMATE COMPOSITION OF REISHI MUSHROOM (*Ganoderma lucidum*) GROWN ON DIFFERENT SAWDUST SUBSTRATES

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ABSTRACT

The experiment was carried out to find out the effect of different sawdust substrates viz. sawdust of gorjan (Rhizophora apiculata) (T_1), sawdust of segun (Tectona grandis) (T_2), sawdust of rain tree (Samanea saman) (T_3) and sawdust mixture of gorjan, segun and rain tree (T_4) on the proximate composition of Ganoderma lucidum. The moisture and protein content ranged from 68.79 to 72.38 % and 25.50 to 27.00 mg/100g respectively. The highest moisture (72.38 %) and protein (27.00 mg/100g) was observed in T_3 which were lowest in T_2 (68.79 %) and in T_1 (25.50 mg/100g) respectively. T_1 contained the highest percentage of carbohydrate (15.25 mg/100g) and lipid (2.65 mg/100g) while lowest carbohydrate (10.03 mg/100g) and lowest lipid (2.10 mg/100g) was recorded in T_3. The highest amount of crude fiber (56.39 mg/100g) was recorded in T_3 and lowest (52.57 mg/100g) was recorded in T_1 while highest value of ash (4.48 mg/100g) was found in T_4 while it was lowest (3.03 mg/100g) in T_2. The highest amount of nitrogen (4.32 %), phosphorus (0.95 %) and potassium (1.49 %) were measured in T_1 whereas the lowest nitrogen (4.08 %), phosphorus (0.79 %) were measured in T_4 and potassium (1.19 %) was in T_3. The highest amount of calcium (27.50 mg/100g), magnesium (89.00 mg/100g), cupper (1.20 mg/100g), iron (53.50 mg/100g), zinc (14.70 mg/100g) and sodium (20.00 mg/100g) was measured in T_1 and the lowest amount of calcium (26.83 mg/100g), magnesium (86.27 mg/100g), cupper (0.90 mg/100g), iron (50.50 mg/100g), zinc (11.90 mg/100g) and sodium (17.17 mg/100g) was measured in T_4.

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Keywords: - *Ganoderma lucidum, substrate, sawdust, proximate composition*

1. INTRODUCTION

Ganoderma lucidum, is a Woody basidiomycotina mushroom belonging to the family of Ganodermaceae of polyporales. They are commonly named as "Lingzhi'' in china, "Youngzhi'' in Korea, "Reishi'' in Japan, and just "*Ganoderma*'' in USA [30]. It is a large, dark mushroom with a glossy exterior and have a woody texture. The Latin word *lucidus* means "shiny" or "brilliant" and refers to the varnished appearance of the surface of the mushroom. *G. lucidum* is widely used in oriental medicine for longevity and health promotion [19]. It is a well-known Asian herbal remedy with a long and impressive range of applications. Global consumption of *G. lucidum* is high, and a large, increasing series of patented and commercially available products that incorporate *G. lucidum* as an active ingredient are available as food supplements. These include extracts and isolated constituents in various formulations, which are marketed all over the world in the form of capsules, creams, hair tonics, and syrups.

It has also become popular because of its promising properties that might extend life span while increasing vigor and vitality [28]. Traditionaly, *Ganoderma* is highly regarded as an herbal treatment and is claimed to alleviate or cure virtually all diseases [11, 17, 23]. *G. lucidum* has been reported to have a number of pharmacological effect including immunomodulating, antiartherosclerotic, anti-inflammation, analgesic, chemopreventive, anti-tumor, radioprotective, sleep-promoting, antibacterial, antiviral (including anti-HIV), hypolipidemic, anti-fibrotic, hepatoprotective, diabetic, antioxidative and radical-scavenging, anti-aging, hypo-glycemic and anti-ulcer properties [6, 13, 14, 22, 33, 35].

Polysaccharides, peptidoglycans, and triterpenes are three major physiologically active constituents in *G. lucidum* [3, 36]. This mushroom contains polysaccharides, polysaccharide- peptide complex, β-glucans, lectins, organic germanium (Ge), adenosine, triterpenoids, phenols, steroids, amino acids, lignin, mycins, vitamins, nucleotides and nucleosides each having their own outstanding medicinal effects [21, 23]. Most fractions of identified polysaccharides and triterpenes have more than hundred compounds that are potent immune-modulators, antioxidants and /or chemo preventive, tumoricidal and anti-diabetic efficacy as the polysaccharide have potential hypoglycemic and hypolipidemic activities [9].

Mushroom cultivation is the only large-scale biotechnological process that creatively utilises lignocellulosics [29]. Using locally available lignocellulosic substrates to cultivate this mushroom is one solution to transform these inedible wastes into accepted edible biomass of high market, medicinal and nutrient values [31]. Most organic matters containing cellulose, hemicelluloses and lignin can be used as mushroom substrate such as, rice and wheat straw, banana leaves, cottonseed hulls, corncob, sugarcane baggase, sawdust, waste paper, leaves, and so on. The amount of nutrition requirement differs according to mushroom species and types of substrate used, the mushroom yield is also related to chemical and biological composition of the substrate [32]. Moreover, difference in mineral content of mushroom are also depended on substrates used [12]. For the cultivation of most medicinal mushrooms the basic substrate is hardwood sawdust (a mixture of fine and coarse sawdust to ensure good aeration) 75-80%, supplemented with wheat bran 20%, gypsum 1%, sucrose 1%, moisture content 60-65% and pH 5.5-6.5. [7, 29]. *G. lucidum* is the annular mushroom grows in a wide variety of dead or dying trees especially in deciduous trees and less frequently found on coniferous trees [7]. Currently, *G. lucidum* grows on a wide variety of wood log, short wood segment, tree stump, sawdust bag and bottle procedures for commercial production [34]. Sawdust obtained from different tree species with a variety of additive ingredients such as wheat bran, corn meal, cereal grains, mineral additives (mostly in the form of gypsum and/or chalk) and other organic materials is well established as it is more beneficial due to the enriched nutritional environment [12, 20, 25].

In Bangladesh sawdust of different tree species are available and is a suitable substrate for mushroom cultivation but very little works have been carried out to find out the performance of *Ganoderma lucidum* on different sawdust substrates. With its growing popularity, many studies on *G. lucidum* composition, cultivation, and reputed effects are being carried out, and there are data that support its positive health benefits. Nutritional analysis of several mushroom species of different origins had been carried out in many laboratories in the world. But nutritional values of locally cultivated reishi mushrooms (*Ganoderma lucidum*) remain speculative. In this work, the effects of various kinds of sawdust substrate medium on the nutritional composition of *G. lucidum* were studied. The aim of the study was to determine the best kind of sawdust and their mixture for cultivation of *G. lucidum* with best nutrient composition.

2. MATERIALS AND METHODS

2.1 Location

The experiment was carried out at Biochemistry laboratory of Sher-e-Bangla Agricultural University and the Tissue Culture Laboratory and culture house of Mushroom Development Institute, Savar, Dhaka, during the period from January to June 2018.

2.2 Experimental materials

Reishi mushroom were grown on different sawdust substrates of 500 g size with supplement. The experiment was carried out with the following treatments:

- T_1= Sawdust of Gorjan (*Rhizophora apiculata*)
- T_2= Sawdust of Segun (*Tectona grandis*)
- T_3= Sawdust of Raintree (*Samanea saman*)
- T_4= Mixture of Sawdust of Gorjan, Segun and Raintree

2.3 Experimental design

The experiment was laid out in Completely Randomized Design (CRD) with three replications to achieve the desired objectives.

2.4 Preparation of spawn packets

Spawn packets of different sawdust were prepared separately. Sawdust was used as a main substrate and wheat bran was used as supplement. For each 500gm spawn packet, 116.7gm sawdust, 58.23gm wheat bran and 1g $CaCO_3$ were mixed and moisture was adjusted at 65% by adding water. The mixture was filled into heat tolerant polypropylene bags of 7" x 10" size and their mouth were plugged by inserting water absorbing cotton and covered with brown paper and tied with a rubber band. Then the previously prepared bags were autoclaved at 121°C and 15 PSI for 1 hour. Each spawn packet was inoculated with the mother culture at the rate of two tea spoon full per packet. Bags were then incubated for mycelium running in the mycelium culture house at 25°C temperature. After 25 days of inoculation, when colonization was complete, the spawn packet was taken to the culture house.

2.5 Spawn packet culture in culture house

The spawn packets with complete mycelium were transferred to the culture house and the brown paper, rubber bands, cotton plug and plastic neck of the spawn packets were removed and the polypropylene bags were wrapped and tied with rubber bands. The plastic bags were opened by 'D' shaped cut on the shoulder side and the sheet was removed. The opened surface of substrate was scraped slightly with a blade for removing the thin whitish mycelial layer. The spawn packets then were then soaked in water for 15 minutes and kept for few minutes to remove excess water. The packets were placed separately side by side on the rack in the culture house.

2.6 Cultural condition

The relative humidity was 80-90% and the temperature was maximum 29.5°C and minimum 23°C. The relative humidity (RH %) and the temperature were maintained by watering thrice daily. Diffused day light and proper ventilation in culture house were maintained for fruiting body development.

2.7 Moisture Analysis

The moisture of fresh mushrooms was determined by using automatic moisture analyzer.

2.8 Determination of total protein

5 g of grinded mushroom was taken with 50 ml of 0.1 N NaOH and boiled for 30 minutes. The solution was cooled in room temperature and centrifuged at 1000 rpm by a table centrifuge machine (*DIGISYSTEM: DSC-200T; Taiwan*) .The supernatant was collected and total protein content was measured according to the Biuret method [26].

2.9 Determination of total Lipid

Total lipid was determined by slight modified method of Folch et al. (1957) [8]. Five gram of grind mushroom was suspended in 50ml of chloroform: methanol (2:1 v/v) mixture then mixed thoroughly and let stand for 3 days. The solution was filtrated and centrifuged at 1000 rpm by a table centrifuge machine. The upper layer of methanol was removed by Pasteur pipette and chloroform was evaporated by heating. The remaining was the crude lipid.

2.10 Determination of crude fiber

10g of Moisture and fat free sample was treated with 0.255N H_2SO_4 and 0.313N NaOH followed by washing with some alcohol and ether. Then transferred to a crucible, dried overnight at 80-100°C and weighed (We) in an electric balance. The crucible was heated in a muffle furnace at 600°C for 5-6 hours, cooled and weighed again (Wa). The difference in the weights (We-Wa) represents the weight of crude fiber.

Crude Fiber (g/100g sample) = [100-(moisture+ fat)] x (We-Wa)/Weight of sample [26].

2.11 Determination of total Ash

1g of the sample was weighed accurately into a crucible and placed on a clay pipe triangle and heated first over a low flame till all the material was completely charred, followed by heating in a muffle furnace for about 5-6 hours at 600 °C. It was then cooled in a dessicator and weighed. Ash content (g/100gsample) = Weight of ash x 100/Weight of sample taken [26].

2.12 Total carbohydrate Estimation

The content of the available carbohydrate was determined by the following equation:

Carbohydrate (g/100g sample) = [100-(Total fat + Total protein + Ash + Crude fiber)] [26].

2.13 Mineral Analysis

Determination of Ca, Mg, Na, K, Fe, Zn, Cu, N and P: The supplied sample was digested with nitric acid to release of Ca, Mg, Na, K, Fe, Zn, Cu, and P. Ca, Mg, Fe, Zn and Cu were determined by atomic absorption spectrophotometry, K was determined by flame photometry, and P was determined by spectrophotometry. The estimation of nitrogen is done by Kjeldhal method [2].

2.14 Determination of metabolizable energy content

Metabolizable energy was calculated by the following formula:

Metabolic Energy (Kcal/100g) = [(3.5 X % Crude Protein) + (8.5 X % Crude Fat) + (3.5 X % Nitrogen Free Extract (Carbohydrate))] [4].

2.15 Statistical analysis

The collected data were analyzed statistically following completely randomized design by MSTAT-C computer package programme. The treatment means were compared by Least Significance Differences (LSD), Duncan's Multiple Range Test (DMRT) [10].

3. RESULTS AND DISCUSSIONS

The experiment was conducted to study the effect of different sawdust substrates on the proximate composition of reishi mushroom. Data of the different parameters analyzed statistically and the results have been presented in the Tables and Figures and discussed under the following headings.

3.1 Effect on moisture content

The moisture content of the fresh fruiting body ranged from 68.79 to 72.38 %. The highest moisture percent was observed in T_3 (72.38 %) and the lowest moisture was in T_2 (68.79 %) which was statistically similar with other treatments (Table-1). The results of the present study is supported by Khan et al. (2009) who found 70.20% moisture in reishi mushroom grown on mixed sawdust. Chang and Miles (1989) reported that moisture content of most edible mushrooms ranges from 70.00 to 94.00 % and for tough edible mushroom it is 50.00 to 75.00 %.

3.2 Effect on protein content

Significant variation was observed on the protein content due to different sawdust substrates. The protein content varied from 25.50 to 27.00 g/100g in the mushroom grown on different sawdust substrates. The highest content of protein was found in treatment T_3 (27.00 g/100g) and the lowest protein was found in T_1 (25.50 g/100g) (Table-1). The results of the present study is supported by Khan et al. (2009) who found 26.40 g/100g protein in reishi mushroom grown on mixed sawdust. Kulsum et al. (2009) found 31.30% protein in sawdust supplemented with cow dung @ 10% in oyster mushroom.

3.3 Effect on carbohydrate content

The highest carbohydrate content was recorded in T_1 (15.25 g/100g) and the lowest carbohydrate was recorded under treatment T_3 (10.03 g/100g (Table-1). The findings of the present study matches with the study of Kulsum et al. (2009) who found that carbohydrate content was ranged from 32.85 to 56.38 % due to sawdust supplemented with different levels of cow dung in oyster mushroom. Khan et al. (2009) found 12.80 g/100g carbohydrates in reishi mushroom grown on mixed sawdust. Alam et al. (2007) found 39.82 to 42.83% carbohydrates in *Pleurotus spp.*

Table-1: Effect of different sawdust substrates on the moisture, protein, carbohydrate, lipid, crude fiber and ash content of *Ganoderma lucidum*

Treatments	Moisture (%)	Dry weight basis (g/100g of mushroom)				
		Protein	**Carbohydrate**	**Lipid**	**Crude fiber**	**Ash**
T_1	69.96 b	25.50 b	15.25 a	2.65 a	52.57 b	4.03 a
T_2	68.79 b	26.76 a	13.72 b	2.12 a	54.37 ab	3.03 b
T_3	72.38 a	27.00 a	10.03 d	2.10 a	56.39 a	4.48 a
T_4	70.30 b	26.20 ab	11.84 c	2.37 a	56.29 a	3.30 b
$LSD_{(0.05)}$	2.00	1.00	1.00	0.89	3.25	0.50
CV (%)	6.51	4.40	4.53	5.65	5.96	6.74

Table in a column followed by a common letter do not differ significantly at 5% level by DMRT. T_1 = Sawdust of Gorjan (*Rhizophora apiculata*), T_2 = Sawdust of Segun (*Tectona grandis*), T_3 = Sawdust of Raintree (*Samanea saman*) and T_4 = Mixture of these Sawdust.

3.4 Effect on lipid content

Reishi mushroom grown on different sawdust substrates showed insignificant difference in lipid content. The lowest lipid content was recorded in T_3 (2.10 g/100g), and it was highest in T_1 (2.65 g/100g) (Table 1). The result of the present study agreed with the findings of Khan et al. (2009) who found 2.7 g/100g lipid in reishi mushroom. Alam et al. (2007) and Kulsum et al. (2009) also reported 4.30 to 4.41% and 3.44 to 5.43% lipids in oyster mushroom grown on different substrates respectively.

3.5 Effect on crude fiber content

Significant variation was found in crude fiber content due to different sawdust substrates. The highest amount of crude fiber was recorded in T_3 (56.39 g/100g) and it was lowest in T_1 (52.57 g/100g) (Table 1). The findings of the present study is supported by Khan et al. (2009) who found 51.50 g/100g crude fiber in reishi mushroom. Chang and Miles (1989) and Kaul (2001) reported that most medicinal mushroom contains high fiber. Kulsum et al. (2009)

found that crude fiber content was ranged from 20.31 to 24.01% in sawdust supplemented with different levels of cow dung.

3.6 Effect on ash content

Significant variation was observed in ash content on different sawdust substrates (Table 1). The highest percentage of ash was observed in the treatment T_3 (4.48 g/100g) and the lowest amount of ash was in T_2 (3.03 g/100g). The result is more or less similar with the findings of Khan et al. (2009) who found 6.50 g/100g ash in reishi mushroom grown on mixed sawdust. Kulsum et al. (2009) who found that ash content was ranged from 6.58 to 8.41% due to sawdust supplemented with different levels of cow dung. Alam et al. (2007) reported 8.28 to 9.02% ash in *Pleurotus spp.*

3.7 Effect on Nitrogen, Phosphorus and Potassium content

Nitrogen content of reishi mushroom significantly influenced by different sawdust substrates (Figure 1). The highest percentage of nitrogen was measured in T_3 (4.32 %) and the lowest nitrogen was in under T_1 (4.08 %). The findings of the present study matches with the study of Moni et al. (2004) who found 4.22 to 5.59 % nitrogen on dry matter basis in fruiting bodies of oyster mushroom. Kulsum et al. (2009) found that nitrogen content was ranged from 1.81 to 5.01% due to sawdust supplemented with different levels of cow dung. Phosphorus content of reishi mushroom was not significantly influenced by different sawdust substrates. The highest percentage of phosphorus was measured in T_1 (0.95 %) and the lowest was in T_4 (0.79 %). The results of the present study agreed with Khan et al. (2009) who found 685mg/100g i.e. 0.69 % phosphorus in reishi mushroom grown on mixed sawdust. Sarker et al. (2007) found 0.97% phosphorus in oyster mushroom grown on sawdust based substrates. Kulsum et al. (2009) also found that phosphorus content was ranged from 0.84 to 0.92 % due to sawdust supplemented with different levels of cow dung. Potassium content of reishi mushroom was significantly influenced by different sawdust substrates. The highest percentage of potassium (1.49) was measured in T_1, which was statistically similar with T_2 and T_4 .The lowest potassium percentage was measured in T_3 (1.19) (Figure 1). The findings of the present study confirms by the study of Chang et al. (1981) who reported that the fruiting bodies of *Pleurotus* contained 1.432 to 1.88 mg/g of K on dry weight of fruiting bodies. Sarker et al. (2007) also found 1.3% potassium in reishi mushroom grown on sawdust based substrates.

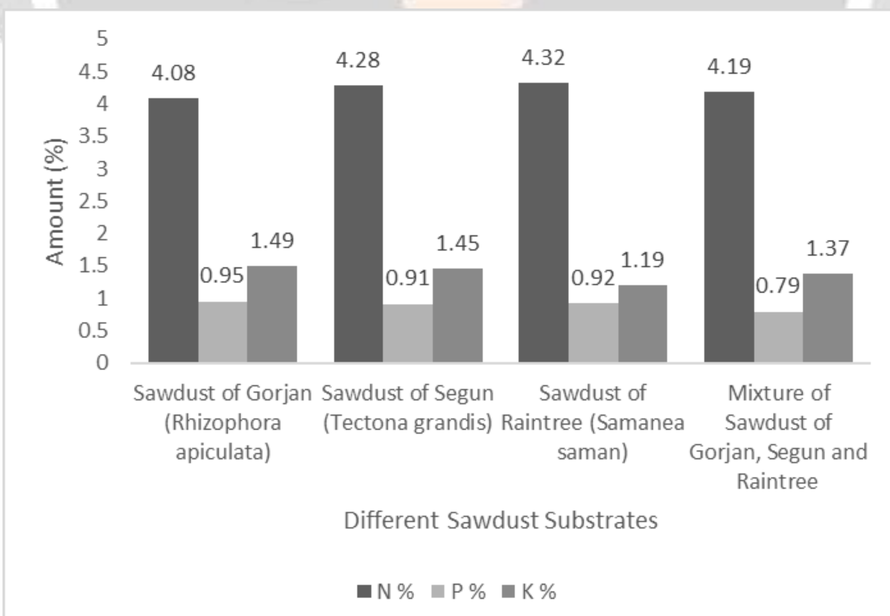

Fig -1: N, P and K contents of *Ganoderma lucidum* grown on different sawdust substrates

3.8 Effect on Calcium, Magnesium and Sodium content

The highest amount of calcium was measured in T_1 (27.50 mg/100g) and the lowest amount was measured in T_2 (26.83 mg/100g) (Table-2). The findings of the present study matches with the study of Alam et al. (2007) who found 22.15 to 33.70 mg/100 g of calcium in reishi mushroom on different sawdust. Khan et al. (2009) found 27.5 mg/100g calcium in reishi mushroom grown on mixed sawdust. Effect on sodium was significantly influenced by different sawdust substrates in reishi mushroom. The highest amount of sodium was measured under in T_1 (20.00 mg/100g) and it was lowest in T_4 (17.17 mg/100g) Magnesium content was significantly influenced by different sawdust substrates in reishi mushroom. The highest amount of magnesium was measured in T_1 (19.00 mg/100g) and the lowest amount was measured in T_4 (16.27 mg/100g) (Table-2). The findings of the present study was supported by Khan et al. (2009) who found 16.80 mg/100g magnesium in reishi mushroom grown on mixed sawdust. Alam et al. (2007) found that magnesium content ranged from 13.4 to 20.22 mg/100g in different oyster mushroom varieties.

3.9 Effect on Cupper, Iron and Zinc content

Cupper, iron and zinc content was significantly influenced by different sawdust substrates in reishi mushroom. The highest amount of cupper (1.20 mg/100g) in T_1, and lowest amount was measured in T_4 (0.90 mg/100g) (Table 2). The highest amount of iron was measured in T_1 (53.50 mg/100g) and the lowest amount was measured under T_4 (50.50 mg/100g) (Table-2). The highest amount of zinc was recorded in T_1 (14.70 mg/100g) and the lowest amount was recorded in T_4 (11.90 mg/100g), (Table-2). The findings of the present study was supported by Khan et al. (2009) who found 53.50 mg/100g iron and 14.70 mg/100g zinc in reishi mushroom grown on mixed sawdust. Alam et al. (2007) reported that zinc and iron content ranged from 16.00 to 20.90 mg/100g and 33.45 to 43.20 mg/100g respectivly in different oyster mushroom varieties. Sarker et al. (2007) found 30.92ppm zinc and 92.09 ppm to 118.40 ppm iron in oyster mushroom grown on sawdust.

Table – 2: Effect of different sawdust substrates on the mineral contents of *Ganoderma lucidum* (mg/100g)

Treatment	Ca	Mg	Cu	Fe	Zn	Na
T_1	27.50 a	19.00 a	1.20 a	53.50 a	14.70 a	20.00 a
T_2	27.37 a	18.23 b	1.10 b	52.50 b	13.90 b	19.15 b
T_3	27.27 a	17.23 c	1.00 c	51.50 c	12.90 c	18.27 c
T_4	26.83 a	16.27 d	0.90 d	50.50 d	11.90 d	17.17 d
$LSD_{(0.05)}$	1.00	0.43	0.02	0.06	0.74	0.29
CV (%)	5.83	5.24	5.65	7.65	6.85	8.65

Table in a column followed by a common letter do not differ significantly at 5% level by DMRT. T_1 = Sawdust of Gorjan (*Rhizophora apiculata*), T_2 = Sawdust of Segun (*Tectona grandis*), T_3 = Sawdust of Raintree (*Samanea saman*) and T_4 = Mixture of these Sawdust.

3.10 Metabolizable energy (Kcal/100g)

The metabolizable energy content of reishi mushroom for different sawdust substrates ranged from 147.46 to 165.15 Kcal/100g (dry weight basis). Highest metabolizable energy was obtained from T_1 (165.15 Kcal/100g) and it was lowest in T_3 (147.46 Kcal/100g) (Fig-2). The findings of the present study agreed with Khan et al. (2009) who reported that metabolizable energy obtained from reishi mushroom grown on mixed sawdust was lower and it was 152.50 Kcal/100g.

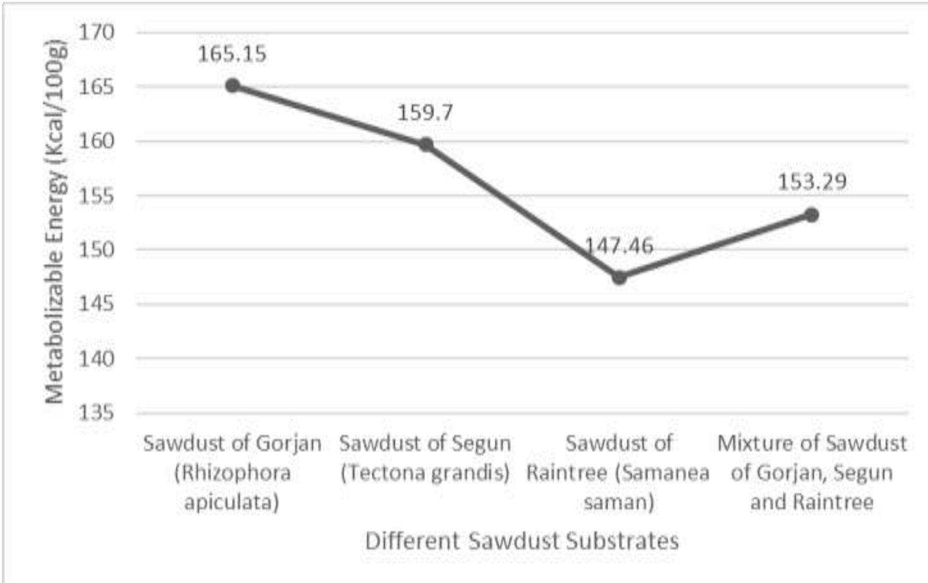

Fig- 2: Metabolizable energy contents of *Ganoderma lucidum* grown on different sawdust substrates

4. CONCLUSIONS

From the present study we can concluded that the reishi mushrooms grown on the sawdust of raintree found best in terms of high protein, fiber and ash with low lipid or fat and carbohydrate content. This mushroom contain high phosphorous and potassium than the mushrooms originated from other sawdust substrates. Therefore, these nutrient content made this mushroom as a low energy healthy foodstuff. So, raintree sawdust can be recommended as a suitable substrates for the cultivation of *Ganoderma lucidum* with best nutrient composition. However, further studies need to be carried out taking more options to justify the present studies.

5. ACKNOWLEDGEMENT

We are thankful to all of Mushroom Development Institute, Savar, Dhaka, for their kind support during this experimental study.

6. REFERENCES

1. Alam, N., Khan, A., Hossain, M. S., Amin S. M. R. and Khan, L. A. (2007). Nutritional Analysis of dietary Mushroom *Pleurotus florida* Eger *and Pleurotus sajorcaj (*Fr.) Singer. *Bangladesh J. Mushroom*. **1(2)**: 1-7.

2. AOAC (2010). Official methods of analysis. Association of official analytical chemists, Washington, DC.

3. Boh, B., Berovic, M., Zhang, J., Zhi-Bin, L. (2007). *Ganoderma lucidum* and its pharmaceutically active compounds. Biotechnol. Annu. Rev.; **13**: 265–301.

4. Chang, S. T. and Miles, P. G. (1989). Edible mushrooms and their cultivation. CRC Press, Boca Raton. pp.1-38.

5. Chang, S. T., Lau, O. W. and Chowdhury, K. Y. (1981). The cultivation and nutritional value of *Pleurotus sajor cuju*. *Eur. J. Appl. Microbiol. Biotechnol.* **12(1)**: 58-62.

6. Chang, S. T. and Buswell, J. A. (1999): *Ganoderma lucidum* (curt.;Fr) P. Karst (Aphylloromycetideae) – a medicinal mushroom. *J. Med .Mushrooms.* **1(2)**: 139-146.

7. Chen, A. W. (1999). A practical guide for synthetic-log cultivation of medicinal mushroom *Grifola frondosa* (Dich. Fr) S.Fr. Gray (Maitake). *Int. J. Med. Mush*. **1**:153-168.

8. Folch, J., Lees, M. & Sloane-stanely, G. H. (1957). A simple method for the isolation and purification of total lipids from animal tissues. *J. Biol. Chem,* **226**: 497-509.

9. Gao, Y., Lan, J., Dai, X., Ye, J. and Zhou, S. H. (2004): A Phase I/II study of Ling Zhi mushroom *Ganoderma lucidum* (W.Curt,: Fr) Loyd (Aphyllophoro mycetideae) extract in patients with type II diabetes mellitus. *J. Med. Mushrooms*. **6(1)**: 33-39.

10. Gomez, K. A. & Gomez, A. A. (1984). Statistical Procedures of Agricultural Research. John wiley and Sons. Inc. New York. pp. 304-307.

11. Hawksworth, D. L. (2001): Mushrooms: the extent of the unexplored potential. *Int. J. Med. Mushrooms*. **3**:333-337.

12. Hoa, H. T., Wang, C. L. and Wang, C. H. (2015). The effects of different substrates on the growth, yield, and nutritional composition of two oyster mushrooms (*Pleurotus ostreatus* and *Pleurotus cystidiosus*). *Mycobiology*. **43**:423–434.

13. Hobbs, C. H. (1995): Medicinal mushrooms: An Exploration of traditional, healing and culture, 2^{nd}, Botanical press, Inc.; santacruz, USA, pp. 252.

14. Jong, S. C. and Birmingham, J. M. (1992): Medicinal benefits of the mushroom *Ganoderma*. *Adv. Appl. Microbiol*.. **37**:101-134.

15. Kaul, T. N. (2001). Biology and conservation of mushrooms. Oxford and IBH publishing co. pvt. Ltd. New Delhi. India. Pp. 117-145.

16. Khan, M. A., Khan, L. A., Hossain, M. S., Tania, M. and Uddin, M. N. (2009). Investigation on the nutritional composition of common edible and medicinal mushrooms cultivated in Bangladesh. *Bangladesh J. Mushroom.* **3(1)**: 21-28.

17. Kingston, D. G. and Newman, D. J. (2005): The search for novel drug lead for predominately antitumor therapies by utilizing Mother Nature’s pharmacophoric libraries. Curr. Opin. Drug. Discov. Devel., **8**:207-227.

18. Kulsum, U., Hoque, S. and Ahmed, K. U. (2009). Effect of different levels of cow dung with sawdust on yield and proximate composition of oyster mushroom (*pleurotus ostreatus*). *Bangladesh J. Mushroom.* **3**(2): 25-31.

19. Leskosek-Cukalovic, Despotovic, S., Lakic, N., Niksic, H. and Tesevic, V. (2010): *Ganoderma lucidum* – Medicinal mushroom as a raw material for beer with enhanced functional properties, *Food Research international.* **43:** 2262-2269.

20. Liang, C. H., Wu, C.Y., Lu, P. L., Kuo, Y. C. and Liang, Z. C. (2016). Biological efficiency and nutritional value of the culinary-medicinal mushroom *Auricularia* cultivated on a sawdust basal substrate supplemented with different proportions of grass plants. *Saudi J Biol Sci.* **8(3)**: 344-350.

21. Liu, G.T. (1999): Recent advanced in research of pharmacology and clinical applications of *Ganoderma lucidum* (Curtis:Fr) , P. Karst . Species (Aphyllophomycetideae) in China. *Int. J. Med. Mushrooms*. **1(1)**: 63-68.

22. McKenna, D. J., Jones, K. and Hughes, K. (2002): Reishi Botanical Medicinal. The desk reference for major Herbal supplements, 2nd; The Haworth herbal press: New York, London, Oxford. Pp. 825-855.

23. Mizuno, T. (1995). *Ganoderma lucidum* and *Ganoderma tsugae:* bioactive substance and medicinal effects. *Food Rev. Int.* **11(1)**: 151-166.

24. Moni, K. H., Ramabardan, R. and Eswaran, A. (2004). Studies on some physiological, cultural and postharvest aspects of Oyster mushroom *Pleurotus ostreatus* (Berk). *Trop. Agril. Res*. **12**: 360-374.

25. Owaid, M. N., Al-Saeedi, S.S., Sabaratnam, V., Al-Assaffii, I. A. and Raman, J. (2015). Growth performance and cultivation of four oyster mushroom species on sawdust and rice bran substrates. *J. Adv. Biotechnol.* **4**:423–429.

26. Raghuramulu, N., Madhaban, N. K. & Kalyanasundaram, S. (2003). A manual of Laboratory Techniques. National Institute of Nutrition. Indian Council of Medical research, pp: 56-58. Hyderabad, India.

27. Sarker, N.C., Hossain, M. M., Sultana, N., Mian, I. H., Karim, A. J. M. S. and Amin, S. M. R. (2007). Impact of different Substrates on Nutrient Content of *Pleurotus ostreatus* (Jacquin ex Fr.) Kummer. *Bangladesh J. Mushroom.* **1(2)**: 35-38.

28. Shiao, M. S., Lee, K. R., Lin, L. J. and Wang, C. T. (1994): Natural product and biological activities of the Chinese medical fungus, *Ganoderma lucidum* in: Food phytochemicals for cancer prevention II: Teas, spices and Herbs, (HO, C.T, Osawar, T.; Huang, M.T and Rosen, R.T.; Eds.)American chemical society: Washington, *C*.P. 342-354.

29. Staments, P. (2000). Growing Gourmet and Medicinal Mushrooms. 3rdedn. Ten Speed Press, Olimpia, WA.

30. Stamets, P. and Yao, C. D. W. (2002): My Comedicinals, An informational treatise on mushrooms. *Olympia, WA*: MycoMedia Productions, pp.96.

31. Tesfaw, A., Tadesse, A. and Kiros, G. (2015). Optimization of oyster (*Pleurotus ostreatus*) mushroom cultivation using locally available substrates and materials in Debre Berhan, Ethiopia. *J. Appl. Biol. Biotechnol.* **3**: 15–20.

32. Tsegaye, Z. and Tefera, G. (2017). Cultivation of oyster mushroom (Pleurotusostreatus Kumm, 1871) using agro- industrialresidues. *J. Appl. Microb. Res.* **1**: 15-20.9.

33. Wasser S. P. and Weis, A. L. (1997). Medicinal Mushrooms *Ganoderma lucidum* (Curtis: Fr), P.karst; Nevo, E.Eds; Peledfus publ House: Haifa, Israel, pp. 39.

34. Wasser, S. P. (2005). Reishi or Ling Zhi (Ganoderma lucidum). Encyclopedia of Dietary Supplements, pp. 603-622.

35. Zhi-bin LIN and Hui-na ZHANG (2004) Anti-tumor and immunoregulatory activties of *Ganoderma lucidum* and its possible mechanisums. *Acta Pharmacol*, **25(11)**:1387-1395.

36. Zhou, X., Lin, J., Yin, Y., Zhao, J., Sun, X. and Tang, K. (2007). Ganodermataceae: Natural products and their related pharmacological functions. *Am. J. Chin. Med.* **35**: 559–74.

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