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Effect of Oyster Mushroom on the Glycemic Status of Type-2 Diabetic Subjects during Ramadan Fast

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Abstract

Dietary habit of Bangladeshi Muslims totally changes during the holy month of Ramadan which may alter the homeostatic mechanisms of the body especially on the metabolic status. Lots of oily fried items consumed at that period which might be the cause of various health hazards including diabetes. The study was undertaken to investigate the effect of oyster mushroom (*Pleurotus ostreatus*) on the glycemic status of type 2 diabetic subjects. The feeding of 50 grams fresh fried oyster mushroom at the *ifter* time during Ramadan fasting significantly reduced the fasting serum glucose (27.27%) of both male and female diabetic subjects as compared to those of the controls (13.97%). Findings of the study indicate that consumption of *P. ostreatus* improves type 2 diabetes.

Key words: *Pleurotus ostreatus,* Ramadan, *Ifter* Type-2 diabetes, Glycemic status.

INTRODUCTION

Mushrooms are wonderful food which comprises a wide range of fungal world. They have been in use not only for culinary purposes, but also for medicinal purposes, since ages. Mushrooms contain many things that fit the definition of food supplements. They contain a wide variety of bioactive molecules including terpenoids, steroids, phenols, nucleotides and their derivates, glycoproteins and polysaccharides (Borchers *et al.*, 1999 and Mizuno *et al.,* 1995). One kind of mushroom may be richer in one of these constituents while another kind will be richer in another. However, mushrooms are generally similar to each other in special food values.

The Oyster mushroom (*Pleurotus ostreatus*), first cultivated in Germany as a subsistence measure during World War I (Eger *et al.*, 1976), is now grown commercially around the world for food. In Japanese, Korean and Chinese cookery, oyster mushroom is frequently used as a delicacy. Mushroom of *Pleurotus* species are also rich in medicinal values and useful in preventing disease such as hypertension, hypercholesterolemia (Khatun *et al.*, 2007 and Choudhury *et al.*, 2008), hyperglycemia, protection of liver (Choudhury *et al.*, 2009) and different types of cancer (Nayana and Janardhanan, 2000). In China, oyster mushroom is indicated for joint and muscle relaxation (Yang and Jong, 1989).

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Now it is accepted that lowering of the serum cholesterol and lipid levels plays a significant role in the prevention of atherosclerosis and other nutrition-related diseases (Bobek *et al.*, 1998). A number of mushroom species have been reported to exhibit hypocholesterolimic effects. This has been attributed to their high dietary fibre levels (Anderson *et al.*, 1988) and other components such as eritadenine, guanylic acid and ergosterol (Breene, 1990). Mushrooms are low in calories and fat, but have higher protein, (19-35% of dry weight) than most vegetables and are good sources of vitamins, especially the B vitamins including riboflavin, niacin and folate (Jansson and Kutti, 2004).

Diabetes mellitus is a universal health problem affecting human society at all stages of development (Rai and Sohi, 1998). Diabetes mellitus is a relatively common disorder in Bangladesh, which has been defined in the genetically and clinically heterogeneous group of disorders. It is primarily caused by degeneration and inactivation of the β cells of islets of Langerhans. There is a serious defect of carbohydrate, fat and protein metabolism in this disorder (Bhal, 1984).

Ramadan is the ninth month of lunar calendar. It lasts for 29 or 30 days, depending on the sighting of the moon. In this month, the Muslims continue their regular activities but abstain from food, liquids, tobacco, sexual activity and medication (oral, inhaler or injection) from sunrise to sunset. However, the sick, the pregnant and nursing mothers and children are exempt; moreover, if a fasting person becomes ill, he or she is allowed to end the fast in the day. Muslims fast to express their gratitude to God and they believe that fasting improves health. There are many schools of thought worldwide who advise fasting, not on religious basis, but for improving health (Gavrankapetanovic, 1997).

Ramadan directly influences the control of diabetes because of the month-long changes in meal times, types of foods, use of medication and daily lifestyle (Maislos, *et al.*, 2001 and Rankin and Bhopal, 2001). Also traditional medicines such as mushrooms are very useful for treatment of certain health problems. Mushrooms are edible fungi which have been used as an antidiabetic drug since ancient time. Among them oyster mushrooms are very useful in the prevention of diabetes mellitus due to the presence of polysaccharides, low glycemic index and lack of sugar and starch. This study was undertaken with the objective to find out the beneficial effect of *P. ostreatus* as *ifter* item of diabetic patients during Ramadan.

MATERIALS AND METHODS

The present study was conducted during Ramadan period with the aid of Strengthening Mushroom Development Project, National Mushroom Development and Extension Center (NAMDEC), Sobhanbag, Savar, Dhaka in association with the Department of Pharmacy and Biochemistry and Molecular Biology, Jahangirnagar University, Savar, Dhaka.

Definition of diabetic subjects: In this study, the diabetic subjects were defined as the subjects whose 12-14 hours fasting serum glucose level were $\geq$ 7 mmol/L.

Subjects: Total 34 adult subjects were included in the study. They were divided into two groups. In diabetes mellitus mushroom group (DMM), 20 subjects aged (years) from 31-65 irrespective of sex and in diabetes mellitus nonmushroom (DMNM) (which was considered as control) group, 14 subjects aged from 32-62 years inclined to fast in the whole Ramadan were considered.

Selection criteria: Adult onset diabetes (type-2) was diagnosed by proper history. The subjects were clarified about the study and after getting their written consent, they were included. The details history was taken from the subjects which included age, sex, occupation, educational status, marital status, family history and drug history. Patients suffering from kidney disease, liver disease, pulmonary tuberculosis and non fast persons were excluded.

Study design: In the study, previously divided two groups were included. DMNM subjects were studied without mushroom supplementation. Any drugs previously taking by the subjects, was continued. Fifty gram of fresh *P. ostreatus* was ensured for each individual of DMM by the responsible workers daily by home visits or from the research center. The mushrooms were collected from NAMDEC. At the beginning of Ramadan, subjects were evaluated for health status. Anthropometric parameters were recorded and fasting blood sample was collected for analysis. Just after ending of Ramadan the subjects were evaluated and all the investigation procedures were repeated. Serum glucose, urea and creatinine were estimated by semi-auto analyzer (3000 evaluation) using commercially available reagent kits.

Anthropometry: Anthropometric measurements were taken *i.e.* height in cm and weight in kg with the use of a digital machine accessorized with a movable headboard. Participants were shoeless and wore light clothing.

Collection of blood sample: Blood samples were collected from the subjects with all aseptic precautions. 10 ml of venous blood were collected from the median cubital vein by a disposable plastic syringe. The needle was detected from the nozzle and blood was transferred immediately into a dry, clean, glass test tube with a gentle push to avoid hemolysis. The test tubes were kept in slanting position till formation of clot. Centrifuging the blood at 3000 rpm for 5 minutes, serum was separated and supernatant was taken into two plastic test tubes (eppendorf), containing 1 ml in each. All the tests were carried out as early as possible. Whenever there was a delay, the serum samples were stored in the Ultra freeze at -20^0C.

Statistical analysis: The recorded characteristics of the subjects were analyzed by standard statistical methods using computer software, SPSS package programme. Data was presented as means ± SE. Comparisons between baseline characteristics of each group were made by using Student's Paired and Unpaired 't' test. Means were significantly difference at p value < 0.05 at 95% confidence limit.

RESULTS AND DISCUSSION

The demographic and clinical profiles of two groups, diabetes mellitus + mushroom (DMM) and diabetes mellitus without mushroom (DMNM) were studied.

In DMM group, the mean ± SE serum urea (mg/dl) before and after Ramadan was 21.7 ± 4.37 and 26.65 ± 3.74, respectively. No significant mean difference of urea (p = 0.405) was observed. In DMNM group, the mean ± SE serum urea (mg/dl) before and after Ramadan was 20.14 ± 1.09 and 23.35 ± 1.86, respectively. No statistically significant mean difference of urea (p = 0.10) was observed. The comparative mean urea of DMM and DMNM (done by independents sample's 't' test) shows – in pre Ramadan state the mean ± SE serum urea of DMM and DMNM was 21.7 ± 4.37 and 20.14 ± 1.09, respectively. No statistically significant mean difference (p = 0.77) between the two groups observed. In post Ramadan state the mean ± SE serum urea of DMM and DMNM was 26.65 ± 3.74 and 23.35 ± 1.86, respectively. Here also, no statistically significant mean difference (p = 0.494) between the two groups was seen (Fig. 1). These findings suggest that supplementation of mushroom as *ifter* item at Ramadan combined or individually have no effect on serum urea level.

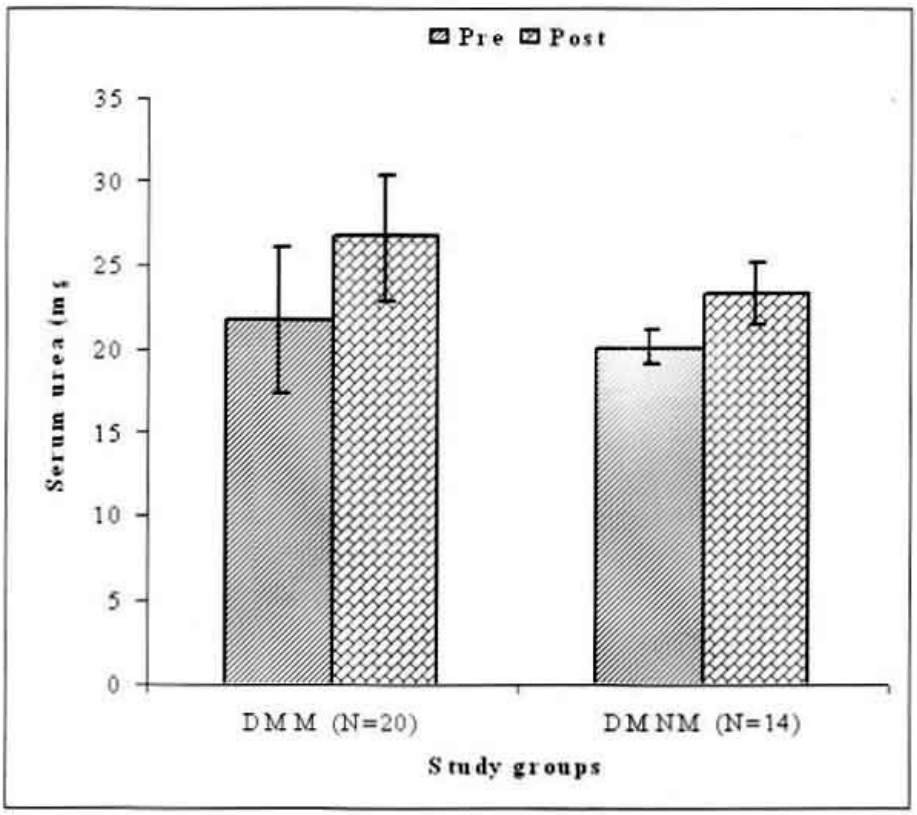

Fig. 1: Mean serum urea of the study groups.

Mean ± SE serum creatinine (mg/dl) in DMM group before and after Ramadan was 0.83 ± 0.13 and 0.84 ± 0.08, respectively. No significant mean difference of creatinine (p = 0.805) observed. In DMNM group, the mean ± SE serum creatinine (mg/dl) before and

after Ramadan was 0.77 ± 0.06 and 0.79 ± 0.05, respectively. No statistically significant mean difference of creatinine (p = 0.62) was observed. The comparative mean creatinine of DMM and DMNM (done by independents sample's 't' test) shows – in pre Ramadan state the mean ± SE serum creatinine of DMM and DMNM was 0.83 ± 0.13 and 0.77 ± 0.06, respectively. No statistically significant mean difference (p = 0.73) between the two groups observed. In post Ramadan state the mean ± SE serum creatinine of DMM and DMNM was 0.84 ± 0.08 and 0.79 ± 0.05, respectively. Here also no statistically significant mean difference (p = 0.64) between the two groups was seen (Fig. 2). These findings suggest that supplementation of mushroom as *ifter* item at Ramadan combined or individually have no effect on serum creatinine level.

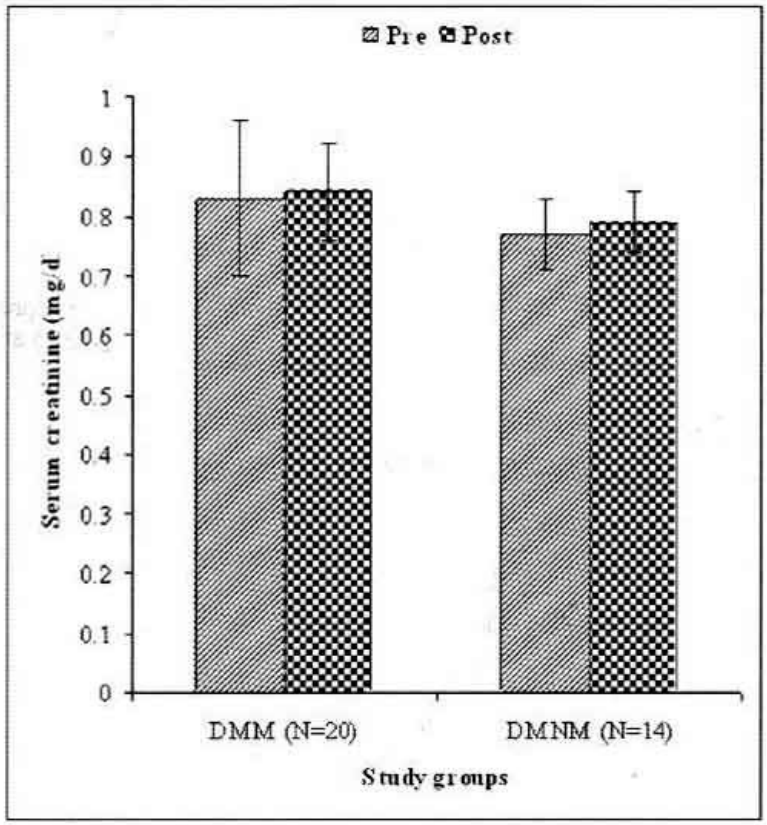

Fig. 2: Mean serum creatinine of the study groups.

In DMM group, the mean ± SE fasting serum glucose (mmol/L) before and after Ramadan was 8.91 ± 0.46 and 6.48 ± 0.57, respectively. A highly significant mean difference of glucose (p = 0.000) observed in pre and post Ramadan state indicating supplementation of oyster mushroom as *ifter* item associated with Ramadan fast significantly reduced serum glucose (27.27%). In DMNM group, the mean ± SE serum glusose level (mmol/L) before and after Ramadan was 8.23 ± 0.40 and 7.08 ± 0.78, respectively. A statistically nonsignificant (p = 0.111) reduction of serum glucose (13.97%) was observed before and after Ramadan (Fig. 3).

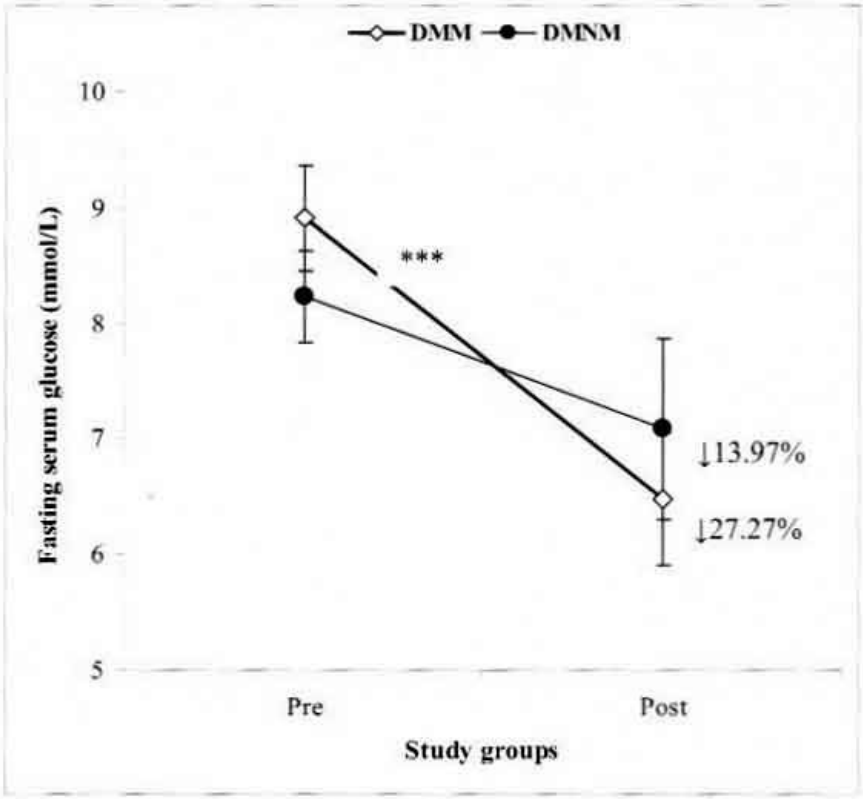

Fig. 3: Mean fasting serum glucose of the study groups (Results are expressed as mean ± SE. Data were analyzed by Pair t test. Means were significantly different at $p<0.05$ at 95% confidence limit).

Considering the obtained findings, it was noticeable that one month consumption of fried *P. ostreatus* as *ifter* item and Ramadan combindly reduced fasting serum glucose of diabetic subjects by 27.27%. On the other hand, only Ramadan fast reduced serum glucose of control group by 13.97%. Thus reducing impact of serum glucose by *P. ostreatus* may be considered as (27.27 – 13.97) 13.3%.

In this study, serum urea and creatinine was estimated to exclude the renal alignment before and after Ramadan period of both mushroom supplemented and non supplemented groups. The obtained finding show that *P. ostreatus* had no effect on serum urea or creatinine level and hence no effect on kidney functions of both of the periods.

Observation of this study focused that supplementation of a considerable amount (50 gram per day) of fried *P. ostreatus* regularly as *ifter* item (1 month) significantly reduced fasting serum glucose in comparison to non mushroom supplemented control group. In different literature it was explained that mushrooms are useful in the prevention of diabetes mellitus due to the presence of polysaccharides and their low glycemic index, lack of sugar and starch. They are nutritive and are richer in protein than cereals, pulses, fruits and vegetables on dry weight (Ghosh, 1990). Due to their low calorific value mushrooms can be consumed by patients with hyperlipidemia (Bano and Rajarathnam, 1982). They are completely devoid of starch and are an excellent inclusion in the diet of diabetic patients. Edible fungi produce secondary metabolites which possess various

therapeutic properties. Mushrooms also contain lots of minerals such as calcium, phosphorous, potassium, iron and copper. They have traditionally been used in the treatment and prevention of diabetes, obesity, heart disease, hyperacidity, constipation, cancer, and hypertension (Suguna, 1995). Oyster mushrooms have been demonstrated to have beneficial effects in animal and human studies individually as well as in combination.

The present study showed that blood sugar was reduced by 27.27% in combined effect of the Ramadan + mushroom (DMM) group, and 13.97% by without mushroom at Ramadan (DMNM) group. There was sufficient effect on glycemic control in both of the groups. The significant fall in fasting blood sugar may be attributed to the hypoglycemic potential of the oyster mushroom supplement. It was reported that oyster mushroom significantly reduced blood glucose level in diabetic subjects (Khatun *et al.*, 2007 and Choudhury *et al.*, 2008). Also reduction in glycated hemoglobin in streptozotocin induced diabetic mice after mushroom supplement was observed (Swanston-Flatt *et al.*, 1989).

Despite the limited size of study population this study was able to demonstrate a significant association between mushroom supplementation and gradual reduction in hyperglycemia in type-2 diabetic subjects. Further studies are needed to verify these observations. In conclusion, the results throw light on the potential use of oyster mushroom for better glycemic control.

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Comparative *in vitro* free radical scavenging effects of *Calocybe indica* and *Pleurotus djamor*

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Abstract

The relative free radical scavenging activities of *Calocybe indica* (Milky) and *Pleurotus djamor* (Pink oyster) extracts were determined against that of the natural antioxidant vitamin C with the use of 2,2-diphenyl-1-picrylhydrazyl (DPPH). The scavenging activities were evaluated both qualitatively by thin layer chromatographic and quantitatively by absorption spectroscopic methods. Total polyphenol contents (TPC) of the mushrooms were determined as gallic acid equivalent (GAE). TPC of the methanol extract of *C. indica* was 0.19 mg and that of the *P. djamor* was 0.2 mg GAE/g of fresh mushroom. The data of the free radical scavenging potentials of *C. indica* and *P. djamor* were subjected to nonlinear regression analyses to calculate IC_{50} (concentrations of mushroom extracts and/or Vit C required to reach half of maximal DPPH free radical inhibition). The analyses revealed IC_{50} for Vit C, of *C. indica* and *P. djamor* extract, respectively, 0.12 mM, 0.06 mM and 0.09 mM GAE.The result obtained from the extract suggest that the anti-free radical effect of *C. indica* was 2-fold higher, as compared to that of the Vit C, while that of the *P. djamor* extract was 1.33-fold higher than that of Vit C. There were no statistical significant differences in the scavenging abilities between milky versus pink oyster, though the former was numerically lower. The result of the present study demonstrates that mushroom extracts are capable of scavenging free radicals.

Key words: *Calocybe indica, Pleurotus djamor* extract, DPPH, Gallic acid, Free radical.

INTRODUCTION

Recently, much attention has been focused on the role of the free radical scavengers in oxidative stress (Sugiyama *et al.*, 1996 and Tsuda *et al.*, 1996). Endogenous antioxidants in plants may play an important role against oxidative damage (Frei *et al.*, 1988 and Naito *et al.*, 1995), and preserved the biological functions of cells (Naito *et al.*, 1994 and Osawa *et al.*, 1990). Free radical oxidative stress has been implicated in the pathogenesis of a wide variety of clinical disorders (Bast *et al.*, 1991).

The antioxidants activity increases interest, particularly in those intended to prevent the presumed deleterious effects of free radicals in the human body, and to prevent the oxidative deterioration of fats and other constituents of food stuffs. In both cases, there is

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a preference for antioxidants from natural rather than from synthetic sources (Abdalla and Roozen, 1999). To protect against the damage caused by such radicals, antioxidants are looked upon with a view of preventing lipid oxidation, disease prevention (vitamin C, vitamin E and carotene), chemo prevention (folic acid and beta-carotene) and health protection (lycopene and ascorbate)[Express Pharma Plus, 1999].There is, therefore, a parallel increase in the use of methods for estimating the efficiency of such substances as antioxidants (Sa'nchez-Moreno, 2002 and Schwarz *et al.*, 2001). One such method that is currently popular is based upon the use of the stable free radical diphenylpicrylhydrazyl (DPPH).

Vitamin C is the major water-soluble antioxidant within the body. The vitamin readily donates electrons to break the chain reaction of lipid peroxidation. The water-soluble properties of vitamin C allow for the quenching of free radicals before they reach the cellular membrane. Tocopherol and glutathione also rely on ascorbic acid (AA) for regeneration back to their active isoforms. The relationship between AA and glutathione is unique. Vitamin C reduces glutathione back to the active form. Once reduced, glutathione will regenerate vitamin C from its dehydroascorbate or oxidized state. Vitamin C has the ability to sequester the singlet oxygen radical, stabilize the hydroxyl radical, and regenerate reduced vitamin E back to the active state. These functions work to halt peroxidation of cellular lipid membranes (Ohkawa,. *et. al.*, 1979).

Bangladesh is blessed with varied agroclimate, abundance of agricultural wastes and manpower, making it most suitable for the cultivation of all the types of temperate, subtropical and tropical mushrooms, so far used in traditional medicine. Milky and pink oyster were reported to ameliorate many diseases, but have remained unexplored in Bangladesh with regard to their medicinal effects on liver injury arising from oxidative stress. The purpose of this study was to examine the basis of this method, and to further examine the use of the parameter "IC_{50}" (equivalent concentration to give 50% effect) which is currently used in the interpretation of experimental data from the method and/or concentrations of mushroom extracts required to reach half of the maximum free radical scavenging potential. Therefore, we used the extracts of an old variety (*C. indica*) and a new variety (*P. djamor*) of available mushrooms in Bangladesh to investigate their relative free radical scavenging activity.

MATERIALS AND METHODS

The strains *C. indica* and *P. djamor* were obtained from National Mushroom Development and Extension Centre (NAMDEC) and the experiment was started in June 2010.

Extraction of *C. indica* and *P. djamor*: Fresh mushrooms were cut into small pieces as much as possible. These were then homogenized with methanol (10%) by Polytron homogenizer. After homogenization the content was subjected to mechanical shaking for 1 hour. Then the content was centrifuged at 1000 g for 15 minutes (Digital centrifuge: DSC-1512SD) at room temperature and filtered through Whatmann No. 1 filter paper.

The residue was re-extracted twice with methanol, as described above. The combined extracts were then transferred into tubes and centrifuged at room temperature at 3000 rpm for 30 minutes. The final extracts were collected, labeled and were stored at -20°C until analysis.

Determination of total phenolic content (TPC): To determine TPC by spectrophotometry, gallic acid was used as standard, according to the method described by the International Organization for Standarization (ISO) 14502-1. For this purpose, 1 ml of the diluted methanolic extract was transferred in triplicate to separate tubes and allowed to dryness at 40 °C. Then 5 ml of a 1/10 dilution of Folin-Ciocalteu's reagent in water was added to each tube and vortexed. Afterwards, 4 ml of a sodium carbonate solution (7.5% w/v) was added and the tubes were allowed to stand at room temperature for 60 minutes before taking absorbance at 765 nm against water. The TPC was expressed as gallic acid equivalents (GAE) in mg/g fresh mushroom. The concentration of total polyphenols in samples was derived from a standard curve of gallic acid ranging from 10 to 50 µg/ml ($r^2 = 0.99$, $P<0.05$).

Qualitative determination of antioxidant properties of *C. indica* and *P. djamor* extracts: The antioxidant activities of the extracts were directly visualized on thin layer chromotagraphic (TLC) plate using 8 µL of 0.4mM DPPH solution. Then after air dry 8 µL of each extract/vitamin C sample was re-subjected to the DPPH-spots and photographed, and intensity was analyzed by Image J after 30 min of incubation.

DPPH radical scavenging activities of *C. indica* and *P. djamor* extracts: Scavenging of the stable radical, DPPH, was assayed in *vitro* by the method of Hatano *et al.* (1988) with slight modification. Aliquots of DPPH solution in ethanol (4 mM, 600 µL) and extract solution of (0 – 0.25mM GAE, 600 µL) were mixed. Butylated hydroxytoluene (BHT) at equimolar concentration was used as positive control. After incubation for 30 min, the absorbance was measured at 517 nm using spectrophotometer (UV-1601PC, Shimadzu, Japan). Antioxidant activity was expressed as the concentration of the extracts (IC_{50}) necessary to scavenge the DPPH-free radicals by 50% that is manifested by the decrease of absorbance. The value was determined graphically by plotting the absorbance data (% inhibition) against the used concentration. To evaluate the antioxidant activity of the extracts, vitamin C was used as common reference standard. Per cent of inhibition was calculated from the following equation:

$$\% \text{ of DPPH scavenging activity} = \frac{(\text{Absorbance of control} - \text{Absorbance of test}) \times 100}{\text{Absorbance control}}$$

Where,
Absorbance of control = Absorbance of DPPH in the absence of extracts,
Absorbance of test = Absorbance of DPPH in the presence of extracts.

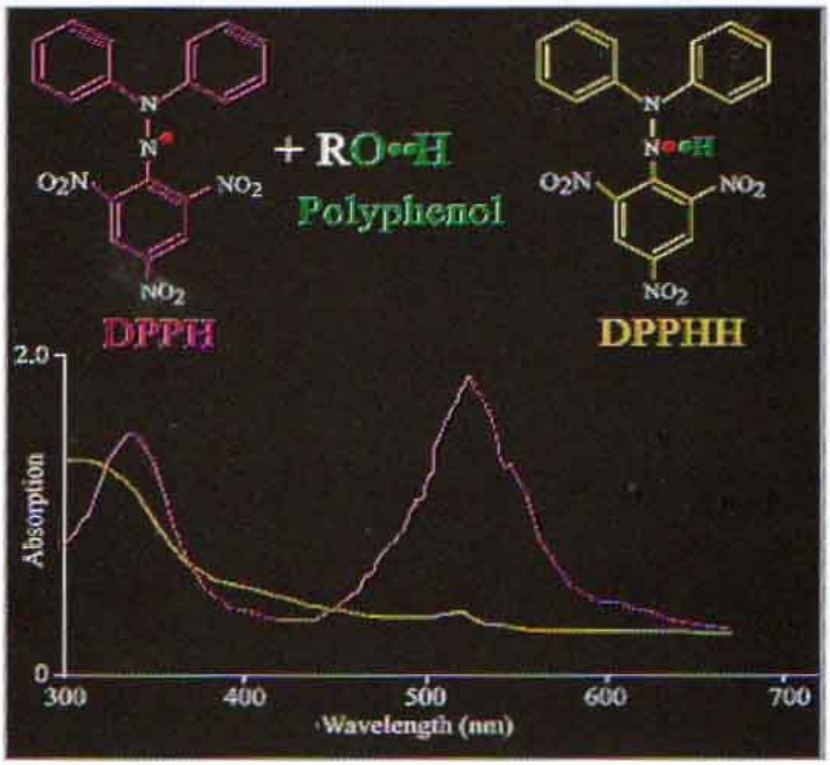

Fig 1. Structure of stable DPPH free radical. The free radical form has characteristic violet to purple color. DPPH has a nitrogen-centered free radical with a characteristic absorption peak at 517 nm. After the free radical is scavenged by the antioxidant, here mushroom extract, its color disappears with the flattening of the peak at 517 nm (Taken from Hossain *et al.*, Oriental Pharmacy and experimental Medicine, *in press*).

Furthermore, to directly visualize the antioxidant activity of the extract, aliquots of 8 µL 4 mM DPPH solution were subjected to thin layer chromatographic (TLC) plate. After air dry, 8 µL of each extract and butylated hydroxytoluene (BHT) and vitamin C was re-subjected to the DPPH spots. After 30 min. of incubation, the spots were photographed and analyzed by Image J.

Statistical Analysis: The results are expressed as mean ± SEM (Standard error of mean). For inter-group differences one-way ANOVA, followed by Fisher's protected least square differences (PLSD) for post hoc comparisons was used. The data for IC_{50} were subjected by nonlinear regression analysis. The statistical programs used were Stat View® 4.01 (Mind Vision Software, Abacus Concepts, Inc., Berkeley, CA, USA) and Graph Pad Prism® (version 4.00; Graph Pad Software Inc., San Diego, CA, USA). A level of <0.05 was considered statistically significant.

RESULTS AND DISCUSSION

In this study, total polyphenol content of *C. indica* was 0.19 mg GAE/g while that of the *P. djamor* it was 0.2 mg GAE/g. The relative DPPH-scavenging activities of the two mushroom extracts versus Vit C are shown in the figure 2. The data of the DPPH-

scavenging activity was fitted to nonlinear regression analysis. The analyses revealed that the IC_{50} value of Vit C was 0.12 mM while that of the *C. indica* and *P. djamor* was 0.06 and 0.09 mM GAE, respectively. The results thus suggest that both of the mushrooms, which contained 0.19~0.2 mg GAE/g fresh tissue, were stronger anti-radicals than vitamin C. According to Folin-Ciocalteu assay (Singleton *et al.*, 1999) milky and pink oyster mushroom extracts possessed a good amount of polyphenols (0.19 mg GAE/g *C. indica* and 0.2 mg GAE/g *P. djamor*) to perform as antioxidant against oxidative species.

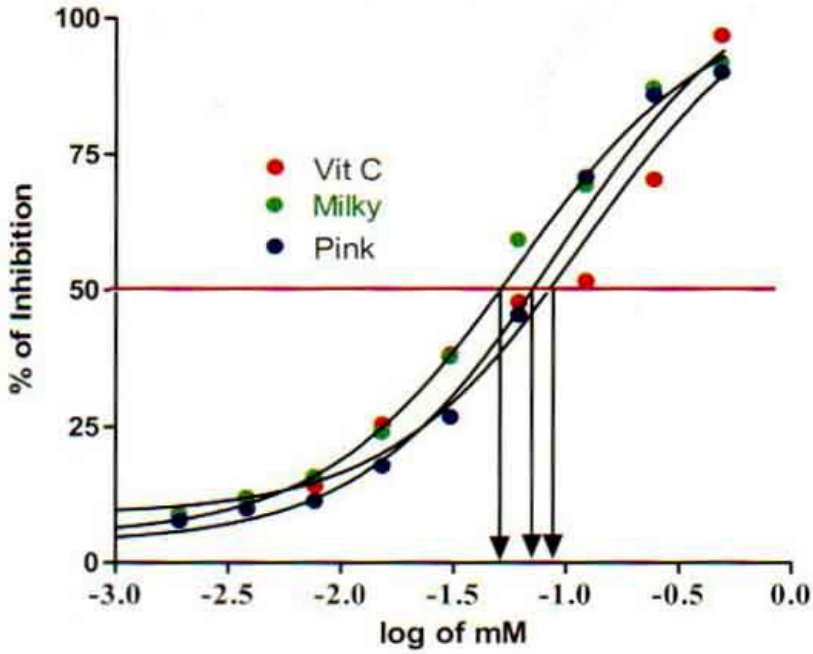

Fig 2: Illustrates the capacity of varying concentrations of mushroom extracts and vitamin C in scavenging the stable DPPH radicals. Data are the representative of triplicate determinations. Data were subjected to nonlinear regression analysis with *top down to bottom, standard slope* equation {Y=Bottom + (TOP-Bottom)/(1+10^((X- logIC$_{50}$)} where IC_{50} is the concentration of antioxidants (here Vit C and mushroom extracts) required to reach half-maximal inhibition.

In this study, the mushroom extracts were able to discolor the characteristic purple color of DPPH (Fig. 3), indicating the qualitative deactivating ability of mushroom extracts of stable free radicals DPPH.

The results were further confirmed by TLC methods, where the characteristic purple color was gradually lightened as a function of concentrations of the mushroom extracts and/or Vit C/BHT (butylated hydroxytoluene, a synthetic antioxidant) (Fig.4). The DPPH scavenging activity of the mushroom extracts in the present investigation was consistent with those of the natural antioxidants in the biological systems such as vitamin C and synthetic antioxidant BHT. The antiradical activity of mushroom extracts was greater in our experimental systems. This suggests that the radical scavenging activities of the extracts are comparable to those of ascorbic acid and or BHT.

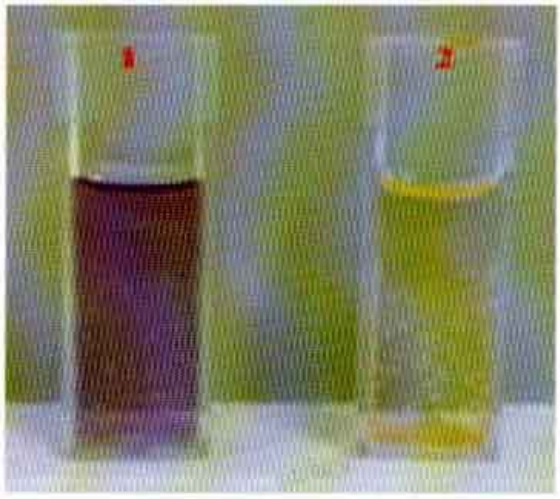

Fig 3. The mushroom extract was added to the cuvette containing the stable free radical DPPH. The purple color (1) disappeared and turned yellow (2).

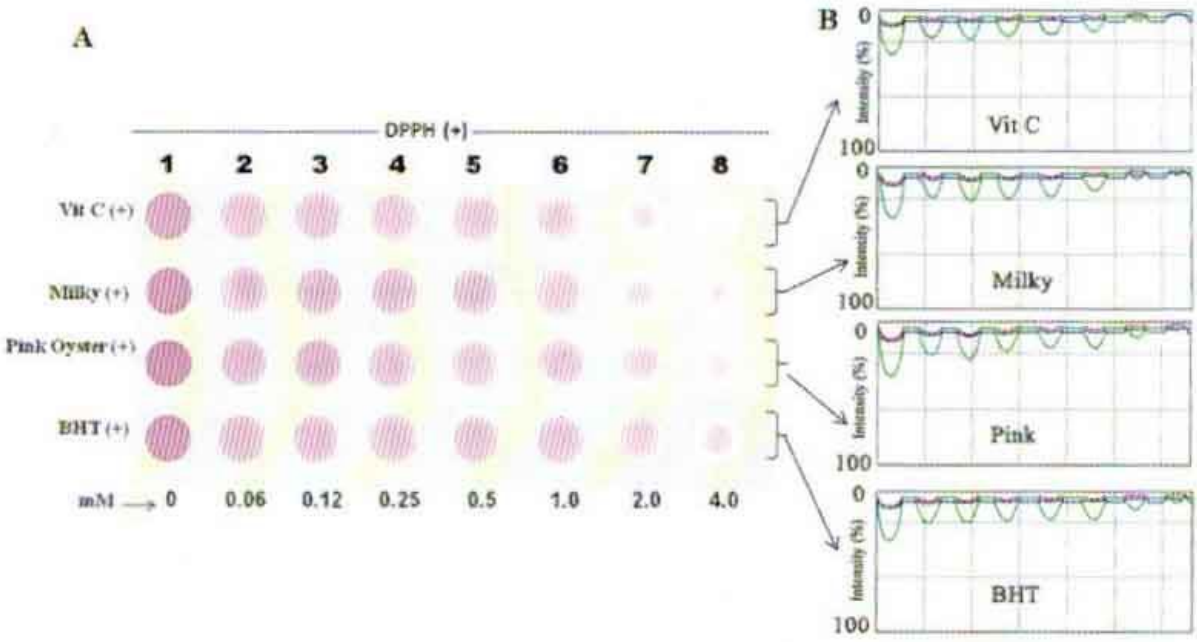

Fig 4. (A) Representative DPPH-stained TLC plate in the absence or presence of *C. indica* and *P. djamor* extract vitamin C and butylated hydroxytoluene (BHT). All clearly scavenged the DPPH free radicals, as indicated by the gradual lightening of the purple color of the spots. The scavenging effects were dose dependent. (B). The intensity (RGB) of the colored spots was digitized, calculated and represented by using NIH ImageJ analyzer.

Finally, liver disease induced by any hepatotoxin and or by increased oxidative stress, lipid peroxidation might be improved with the ingestion of the mushroom extracts or direct consumption of mushrooms. The study might also have an important impact on the individuals of Bangladesh, who are suffering from hepatic dysfunctions and need to

protect the disease by protection against oxidative damage can be advised to consume one of the mushrooms stated in this study along with their daily meal, instead of ingesting high priced natural antioxidants like vitamin E and C. Further extensive study is, indeed, necessary to determine the precise mechanism of actions of mushroom extracts on impaired liver functions originated from oxidative stress.

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Effect of Light Intensity and Its Management Practices on Yield of *Ganoderma lucidum*

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Abstract

Effect of light management practices on yield and yield attributes of reishi mushroom (*Ganoderma lucidum*) in Bangladesh condition were studied. The minimum days required for antler initiation (4.25) and conk formation (7.00) was found at 0 lux of light intensity where the minimum day required for first harvesting (40.00) was found at 570 lux. The highest number of effective fruiting bodies (3.50) was harvested at 80 lux. The highest diameter of pileus (5.50 cm), diameter of stipe (2.82 cm) and length of stipe (9.00 cm) were found at 7 lux but the highest thickness of pileus (1.87 cm) was found at 570 lux. The highest biological yield (35.75 g), economic yield (35.00 g), dry yield (10.13 g) and biological efficiency (17.88 %) were found at 5 lux. The response of different yield parameters to the same or similar light intensities was different due to light management practices, responsible for controlling environmental factors- humidity, temperature and CO_2.

Key Ward: Reishi mushroom, yield, biological efficiency, light intensity.

INTRODUCTION

Reishi (*Ganoderma lucidum*) is one of the best medicinal mushrooms belonging to Polyporaceae (or Ganodermaceae) of Aphyllophorales. Its fruiting body is called "Reishi" in Japanese and "Lingzhi" in Chinese (Wagner, *et al.*, 2003). Lingzhi or Reishi contains various chemical substances, including more than 119 different triterpenes and several types of polysaccharides (Hsieh and Yang, 2004). Pharmaceutically active compounds from *Ganoderma lucidum* include triterpenoids, proteins, steroids, alkaloids, nucleotides, lactones and fatty acids (Habijani, *et al.*, 2001). *Ganoderma lucidum* and its isolates have been known as a traditional remedy, used in Chinese and Japanese traditional medicine for treatment of several diseases, such as hepatitis, hypertension, haepatopathy, gastric ulcer, arteriosclerosis, nephritis, arthritis, neurasthenia leukopenia, diabetes, anorexia insomnia, chronic hepatitis, hyperglycemia, chronic bronchitis, bronchial asthma, cancer and others (Wagner, *et al.*,2003 and Habijani, *et al.*, 2001). The environmental parameters (*e.g.* temperature, light, relative humidity, O_2 and CO_2) are of great importance in obtaining well-formed *Ganoderma* mushrooms (Veena, *et al.*, 2010). Among the environmental factors light is the most important one. For growth and development of the mushroom light has auspicious and adverse effect. According to some reports, mushroom

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requires light for the initiation of fruiting body. But the requirement of light is not same for all stages of fruiting body formation (Chang and Miles, 1993a). Durand and Jacques (1982) expressed their opinion against the light use. They expressed that light may inhibit fruiting body formation. Therefore, this study was aimed to determine the optimum light intensity and its management practices for the highest yield performance of reishi mushroom.

MATERIALS AND METHODS

The study was carried out in the National Mushroom Development and Extension Centre (NAMDEC), Savar, Dhaka from February to July, 2010. Sixteen different light intensities and their management practices, T_1= 15 lux (Covered by a single layer of black cloth and a poly propylene (pp) sheet), T_2= 850 lux (Covered by a single layer of white cloth and a pp sheet), T_3= 10 lux (Covered by a single layer of black cloth and white cloth and a pp sheet), T_4= 40 lux (Covered by a single layer of hessian (Jute chot) and a pp sheet), T_5= 450 lux (Covered by only a pp sheet), T_6 = 0 lux (Covered by a double layer of black cloth and a pp sheet), T_7= 300 lux (Covered by a double layer of white cloth and a pp sheet), T_8 = 7 lux (Covered by a double layer of hessian (Jute chot) and a pp sheet), T_9 = 20 lux (Covered by a single layer of black cloth), T_{10} = 350 lux (Covered by a single layer of white cloth), T_{11} = 12 lux (Covered by a single layer of black cloth and white cloth), T_{12} = 80 lux (Covered by a single layer of hessian (Jute chot), T_{13}= 570 lux (Open, i.e. no covering), T_{14}= 0 lux (Covered by a double layer of black cloth), T_{15}= 330 lux (Covered by a double layer of white cloth) and T_{16}= 5 lux (Covered by a double layer of hessian (jute chot) were considered as treatments.

Preparation and opening of spawn packet and watering in the culture house were done following Hossain *et al.* (2009). The experiment was laid out following completely randomized design (CRD) with 16 treatments and 4 replications. Days required for antler initiation, conk formation and first harvest were counted from opening of the packets. Number of well developed fruiting body was recorded as the number of effective fruiting body. The thickness and diameter of pileus and length and diameter of stipe were measured by a slide calipers. The biological, economical and dry yields per 500g spawn packet were recorded by weighing with an electric balance. The biological efficiency was calculated as percentage of fresh mushrooms in relation to dry weight of substrates.

Data were analyzed following Gomez and Gomez (1984) using MSTAT-c computer program. Means separation were computed following Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Days required for antler initiation: Days required for antler initiation (DRAI) differed significantly and ranged from 4.25 to 8.00 (Table 1). The lowest DRAI (4.25) was observed in treatment T_6 and T_{11} at 0 and 12 lux which was statistically similar to all

other treatments except T_{13} and T_{15}. The highest DRAI (8.00) was found at 570 lux (T_{13}) which was significantly higher to other treatments. Datta and Chakraborty (2002) reported that light enhanced primordia formation.

Days required for conk formation: Days required for conk formation (DRCF) differed significantly and ranged from 7.00 to 13.50 (Table 1). The highest DRCF (13.50) was found in treatment T_7 and T_{13} at 300 and 570 lux, which was statistically similar to treatments T_1, T_4 and T_{15}. The lowest DRCF (7.00) was found at 0 lux (T_1), which is statistically similar to treatments T_9 and T_{12}. From the study it is clear that that low light intensity (near about 0 lux) is suitable for conk formation.

Days required for first harvesting: Days required for first harvesting (DRFH) differed significantly and ranged from 40.00 to 52.00 (Table 1). The highest DRFH (52) was found in treatment T_8 at 7 lux, which was statistically similar to treatments T_4 and T_3. The lowest DRFH (40.00) was found in treatment T_{13} at 570 lux, which was statistically similar to treatments T_5, T_6, T_7, T_{11} and T_{14}.

Number of effective fruiting bodies: The number of effective fruiting bodies (NEFB) differed significantly and ranged from 1.50 to 3.50 (Table 1). The highest NEFB (3.50) were harvested from the treatments T_{16} at 5 lux, which was statistically similar to treatments T_5, T_8, T_{10}, T_{11}, T_{12} and T_{14}. The lowest (1.50) NEFB were harvested from the treatment T_{13} at 570 lux, which was statistically similar to treatments T_1, T_2, T_3, T_7 and T_9. Datta and Chakraborty (2002) reported that light enhanced the production of fruiting bodies.

Diameter of pileus: The diameter of pileus differed significantly and ranged from 3.05 to 5.50 cm (Table 1). The highest diameter of pileus (5.50 cm) was found at 570 lux (T_{13}) (Table 1). The lowest diameter of pileus (3.05 cm) was found at 0 lux (T_{14}). The treatment T_{14}, was statistically similar to others except T_{13}, T_9 and T_8. Chang and Miles (1993b) reported that light inhibited pileus expansion.

Diameter of stipe: The diameter of stipe differed significantly and ranged from 0.72 to 2.8 cm (Table 1). The highest diameter of stipe (2.82 cm) was found at 570 lux (T_{13}) (Table 1) which was significantly higher to other treatments. The lowest diameter of stipe (0.72 cm) was found at 350 lux (T_{10}) which was statistically similar to other treatments except T_{13}. Hossain *et al.* (2009) found that the minimum diameter of stipe (0.95 cm) and the maximum (1.50 cm) when reishi was cultured at light at 250-350 lux. Chang and Miles (1993b) reported that light inhibited stipe expansion.

Length of stipe: The length of stipe differed significantly and ranged from 2.55 to 9.00 cm (Table 1). The 7 lux light intensity was favorable for producing the highest length of stipe (9.00 cm) in T_8 (Table 1), which was statistically similar to treatment T_{16}. The lowest length of stipe (2.55 cm) was found at 20 lux (T_9), which was statistically similar to treatments T_1, T_5, T_{10}, T_{11}, T_{12}, T_{13}, T_{14} and T_{15}. Triratana (1976) observed the elongated stipes in absence of light.

Thickness of pileus. The thickness of pileus differed significantly and ranged from 1.10 to 1.90 cm (Table 1). The 570 lux light was favorable for the highest (1.90 cm) thickness of pileus in T_{13} (Table 1), which was statistically similar to treatments T_1, T_5 and T_{10}. The lowest thickness of pileus (1.10 cm) was found at 7 lux (T_8), which was significantly similar to treatments T_4, T_6, T_9, T_{11}, T_{12}, T_{14} and T_{15}. Hossain *et al.* (2009) found 1.13-1.53 cm thick pileus at the light intensity of 250-350 lux.

Biological yield: Highly significant variation was observed in biological yield (BY) of reishi mushroom among the 16 treatments (Table 1) and ranged from 24.75 g to 35.75 g. The highest BY (35.75 g) was observed in treatment T_{16} at 5 lux, which was statistically similar to treatments T_5, T_8, T_{10}, and T_{14}. The lowest BY (24.75 g) was observed in treatment T_{13} at 570 lux, which was statistically similar to treatments T_1, T_2, T_3, T_7, and T_{12}.

Economic yield: The Economic yield (EY) differed significantly and ranged from 24.38 to 35.00 g. The highest EY (35.00g) was observed in treatment T_{16} at 5 lux, which was statistically similar to treatments T_8, T_{10}, T_{15} and T_{14}. The lowest EY (24.38 g) was observed in treatment T_{13} at 570 lux, which was statistically similar to treatments T_1, T_3, T_7, and T_{12}.

Dry yield: Dry yield (DY) of reishi mushroom was varied significantly and ranged from 10.13 to 6.95 g (Table 1). The highest dry yield (10.13 g) was observed in treatment T_{16} at 5 lux followed by treatments T_5, T_6 and T_{10}. The lowest dry yield (6.92 g) was observed in treatment T_2 at 850 lux, which was statistically similar to treatments T_7, T_8, T_9, T_{11}, T_{13} and T_{14}. Hossain *et al.* (2009) obtained 2.63- 6.13g dry yield at 250-350 lux light.

Biological efficiency: Biological efficiency (BE) of reishi mushroom was worked out against the dry weight of substrate. Significant variation was observed in BE and ranged from 12.38 to17.88% (Table 1). The highest BE (17.88 %) was observed at 5 lux which was statistically similar to T_5, T_8 and T_{10}. The lowest BE (12.38%) was observed at 570 lux.

The results revealed that the response of different yield parameters to the same or similar light intensities was different that might be due to different light management practices which were responsible for controlling other environmental factors like humidity, temperature and CO_2 level.

Table 3. Effect of light intensity and its management practice on yield attributes and yield of reishi mushroom

Treatment	Days to antler initiation	Days to conk formation	Days to first harvest	Number of effective fruiting body	Diameter of pileus (cm)	Diameter of stipe (cm)	Length of stipe (cm)	Thickness of pileus (cm)	Biological yield (g)	Economic yield (g)	Dry yield (g)	Biological efficiency (%)
T_1	5.00 bc	13.25 a	42.00 c	1.50 d	4.17 bc	1.37 b	3.40 de	1.52 ab	24.75 g	24.50 e	7.30 c	12.38 h
T_2	5.75 bc	11.00 cd	42.00 c	2.25 bcd	3.70 bc	0.97 b	4.62 bcd	1.52 b	28.50 d-g	28.50 b-e	6.92 c	14.25 efg
T_3	5.25 bc	12.00 bc	51.75 a	2.00 cd	4.20 bc	1.42 b	6.00 b	1.52 b	29.00 c-g	28.25cde	7.62 c	14.75 defg
T_4	4.75 bc	12.50 ab	51.50 ab	2.50 bc	3.82 bc	1.07 b	5.87 b	1.40 bc	30.50 b-f	29.75 bcd	7.60 c	15.25 cde
T_5	5.25 bc	10.00 de	40.25 d	3.00 ab	3.82 bc	0.95 b	3.50 de	1.50 ab	33.00 a-d	32.50 abc	9.50 ab	16.50 abcd
T_6	4.25 c	8.50 fg	41.00 d	2.50 bc	3.52 bc	0.80 b	2.75 e	1.35 bc	30.00 b-f	30.00 bcd	8.80 abc	15.00 def
T_7	5.75 bc	13.50 a	40.50 d	2.00 cd	3.97 bc	1.45 b	5.50 bc	1.50 b	26.75 efg	25.75 de	7.77 bc	13.25 fgh
T_8	4.75 bc	10.00 de	52.00 a	3.00 ab	4.47 b	1.25 b	9.00 a	1.10 c	34.00 ab	33.00 ab	8.37 bc	17.00 abc
T_9	4.75 bc	8.00 gh	42.50 c	1.75 cd	4.50 b	1.22 b	2.55 e	1.20 bc	30.25 b-f	29.75 bcd	8.25 bc	15.13 de
T_{10}	5.50 bc	10.25 de	42.25 c	3.00 ab	3.55 bc	0.72 b	3.00 de	1.52 ab	33.50 abc	33.00 ab	8.57 abc	17.50 ab
T_{11}	4.25 c	9.25 ef	40.50 d	3.00 ab	3.72 bc	1.17 b	4.00 cde	1.30 bc	30.50 b-f	30.00 bcd	7.95 bc	15.25 cde
T_{12}	5.00 bc	8.00 g	42.50 c	3.50 a	3.55 bc	1.12 b	3.32 de	1.27 bc	26.00 fg	26.00 de	7.20 c	13.00 gh
T_{13}	8.00 a	13.50 a	40.00 d	1.50 d	5.50 a	2.82 a	2.97 de	1.87 a	24.75 g	24.38 e	8.15 bc	12.30 h
T_{14}	5.50 bc	7.00 h	40.00 d	3.00 ab	3.22 c	1.32 b	2.87 de	1.35 bc	31.50 a-e	30.75 abc	8.20 bc	15.75 bcde
T_{15}	6.00 b	13.00 ab	42.00 c	2.50 bc	3.05 c	1.27 b	3.75 de	1.35 bc	30.25 b-f	30.00 bcd	7.55 c	15.13 cde
T_{16}	5.00 bc	10.25 de	50.75 b	3.50 a	3.97 ab	0.95 b	8.25 a	1.50 b	35.75 a	35.00 a	10.13a	18.00 a
CV	16.96	06.79	01.47	20.16	17.50	35.78	24.85	15.94	9.76	9.55	13.61	7.73%

In a column, means followed by a common letter are not significantly different at 5% level by DMRT test. [T_1= 15 lux (Covered by a single layer of black cloth and a poly propylene (pp) sheet); T_2= 850 lux (Covered by a single layer of white cloth and a pp sheet); T_3= 10 lux (Covered by a single layer of black cloth and white cloth and a pp sheet); T_4= 40 lux (Covered by a single layer of hessian (Jute chot) and a pp sheet); T_5= 450 lux (Covered by only a pp sheet); T_6 = 0 lux (Covered by a double layer of black cloth and a pp sheet); T_7= 300 lux (Covered by a double layer of white cloth and a pp sheet); T_8 = 7 lux (Covered by a double layer of hessian (Jute chot) and a pp sheet); T_9 = 20 lux (Covered by a single layer of black cloth); T_{10} = 350 lux (Covered by a single layer of white cloth); T_{11} = 12 lux (Covered by a single layer of black cloth and white cloth); T_{12} = 80 lux (Covered by a single layer of hessian (Jute chot)); T_{13}= 570 lux (Open); T_{14}= 0 lux (Covered by a double layer of black cloth); T_{15}= 330 lux (Covered by a double layer of white cloth); T_{16}= 5 lux (Covered by a double layer of hessian (jute chot)].

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Studies on the age of fruiting bodies for the harvesting of milky mushroom

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Abstract

The sizes of fruiting body of milky mushroom increased with the increases of age up to a certain limit and the appearance of fruit body becomes less attractive with the increases of age over 6 days. The highest number of flushes (4.25) and fruiting body (6.00) was obtained from 4 days old fruiting body. The yield of mushroom increased with the increase of ages of fruiting body up to 10 days and decreased thereafter. The highest yield (219.30 g/packet) was obtained from 10 days old fruiting body which did not differ significantly with 6 and 7 days. The yield decreased with the increases of age of fruiting body over 10 days and the mushroom should be harvested at 6th day after pinhead formation.

Key words: Milky mushroom, yield, biological efficiency, harvesting period.

INTODUCTION

Milky mushroom (*Calocybe indica*), milky white in colour, delicate in texture, robust in size, is one of the most promising mushrooms in Bangladesh. It is also of great choice to the local consumer due to its delicious taste. Its longer shelf life and lucrative market value attract the attention of prospective growers. The size, shape, colour and taste of the mushroom depends on the maturity of fruiting body. If the mushroom is harvested prematurely the yield may be reduced. On the other hand, if it is harvested at over age, the mushroom shades spores and loses its colour. The size of the mushroom become robust and the sporophore become fibrous, leathery and tasteless. The open and over mature mushrooms have low market acceptability. So, it is very important to harvest mushroom in time. In India, the mushroom is harvested at 6-8 days after pinhead formation (Krishnamoorthy and Amutha, 2007). If the crop is harvested within 4-8 days of pin head formation, the mushroom maintains their crop cycle. Otherwise, the next flush will come too late and yield will be reduced (Krishnamoorthy and Amutha, 2007). Therefore, the present study was undertaken to determine the right age of the mushroom for harvesting in Bangladesh condition.

MATERIALS AND METHODS

Twelve different harvesting age of fruiting body was evaluated for milky mushroom harvest at the National Mushroom Development and Extension Centre (NAMDEC), Savar, Dhaka. These are 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14 and 15 days.

Mother culture, a seed material for mushroom cultivation was prepared by following standard procedure (Krishnamoorthy, 1981). For this purpose, uninfested, clean wheat grains were boiled in water till grains become soft but were not allowed to split open. The moisture content of boiled grains was allowed to leave by spreading them on plastic sheet so that the excess water is evaporated to obtain 50 to 55 per cent moisture. Then, the grains were mixed with 0.5 per cent calcium carbonate so that the pH of the grains is around 7.0 to 7.8 and they do not form clumps. This grain was filled into 7" × 10" size polypropylene bags and then sterilized for 1 hour at 120^{0}C. Upon cooling for 24 hours, the content was inoculated with fresh mycelial culture of milky mushroom under aseptic condition. The containers were incubated at 26-28°C for 15-20 days to attain complete ramification over the grains. These seed materials were used freshly for cultivation of aforementioned mushroom.

Rice straw substrate was used for the cultivation of milky mushroom. The straw was chopped to convenient length of about 6 to 10 cm. The substrate was treated with hot water of 60^{0}C for 60 minutes and then allowed to drain out the excess water by hanging the straw bags for 20 hours. The hot water treated straw was spread on a clean cemented floor out side the room to remove excess moisture. Then, the substrate was filled into the polythene bags (9"×12") and seeded with 5% milky mushroom grain spawn by following the method of Krishnamoorthy (1981). These bags were incubated in a culture room. After 18-22 days, the substrate was completely colonized by the mycelium and polythene cover was opened.

Decomposed cow dung and loamy soil (3: 1, v/v) was used as the casing material and was sterilized at 65^{0}C for 4 hours. Casing material was covered over the mycelium on the substrate up to 4 cm thickness. Watering was done at regular interval to maintain moisture at 60 to 70%. Pin head appeared at 14-17 days and developed into fruiting bodies.

The fruiting bodies were harvested according the treatment and the data were collected on the length of stipe, diameter of stipe, diameter of pileus, thickness of pileus, number fruiting body, yield and biological efficiency (BE). The BE was measurd by the formula: BE= Fresh weight of mushroom x 100/ Dry weight of substrate.

The experiment was laid out in Completely Randomized Design (CRD) with 4 replications. The data were statistically analyzed following the CRD with MSTAT-C computer programme. Means were computed following Duncan's Multiple Range Test (DMRT) using the same programme.

RESULTS AND DISCUSSION

Dimension of fruiting body: Highly significant variation was observed in sizes of fruiting body harvested at different ages. The sizes of fruiting body increased with the increases of ages up to a certain limit. The length of stipe was the minimum (3.95 cm) at 4 day old fruiting body and it was increased with the increases of age up to 12 day. No significant variation was observed in stipe length among the fruit bodies of 12 to 14 days.

Diameter of stipe was increased with the increase of age of fruiting body up to 10 day and decreased thereafter. The cause of reduction of stipe diameter might be due to shrinking of fruiting body as the supply of nutrient stopped but respiration continued at older fruit bodies.

The diameter and thickness of pileus increased with the increases of age of fruiting body up to 11 and 12 days respectively. However, significant increase in these two parameters was not observed thereafter.

Table 1. Size of fruiting body at different age

Age of fruiting body (day)	Length of stipe (cm)	Diameter of stipe (cm)	Diameter of pileus (cm)	Thickness of pileus (cm)
4	3.95 h	2.35 f	4.35 g	1.97 g
5	4.32 g	3.15 cd	4.25 g	2.57 de
6	6.17 f	3.52 b	8.52 f	2.27 f
7	6.07 f	3.32 bc	9.40 e	2.32 f
8	6.50 e	3.25 cd	10.30 d	2.37 ef
9	6.65 e	3.87 a	11.50 c	2.57 de
10	9.35 c	4.00 a	12.02 b	2.70 cd
11	9.00 d	3.27 bcd	12.65 a	2.82 bc
12	9.77 ab	3.05 d	11.95 b	3.15 a
13	9.60 b	3.17 cd	11.95 b	3.17 a
14	9.65 b	3.15 cd	12.25 b	3.00 ab
15	9.95 a	2.77 e	12.77 a	3.15 a
CV (%)	2.20	5.18	2.13	5.29

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Appearance of fruit body: The appearance of fruit body becomes less attractive with the increases of age over 6 days (Fig. 1). It loses its beautiful colour and become rusty. The fruiting body also loses its tenderness and delicacy.

a. 4 day old b. 6 day old c. 12 day old

Fig.1. Fruit bodies of different ages of milky mushroom (a. 4-days old; b. 6-days old and c. 12-days old milky mushroom).

Yield attributes and yield: The highest number of flushes (4.25) was observed at 4-days old fruiting body and it did not differ significantly up to 6-days old. The flush number was decreased with the increases of age of fruiting body (Table 2).

The number of fruiting body was significantly influenced by the age (Table 2). The highest number of fruiting body was obtained from 4-days old fruiting body which was significantly higher than all other ages except 5-days old. The lowest number of fruiting body was obtained from 15 days old fruiting body.

The yield of mushroom increased with the increase of ages of fruiting body up to 6-days and decreased thereafter. The highest yield (219.30 g/packet) was obtained from 10 days old fruiting body which did not differ significantly with 6 and 7-days old. The yield decreased with the increases of age of fruiting body over 10-days (Table 2). The cause of lower yield at higher aged fruit body might be due to over staying of fruit bodies on bed delays the next crop resulting low yield.

Table 2. Effect of age of fruiting body on yield attributes and yields of milky mushroom

Age of fruiting body (day)	Number of flushes	Number of fruiting (body/packet)	Yield (g/packet)
4	4.25 a	6.00 a	175.3 d
5	3.75 ab	5.50 ab	174.8 d
6	3.75 ab	4.75 bc	215.0 ab
7	3.50 bc	4.50 cd	215.3 ab
8	3.50 bc	2.50 e	209.8 b
9	3.25 bcd	2.75 e	210.5 b
10	3.00 cde	3.75 d	219.3 a
11	3.00 cde	2.50 c	184.0 c
12	2.75 def	2.75 e	177.8 d
13	2.50 efg	2.25 e	145.0 f
14	2.25 fg	2.50 e	158.8 e
15	2.00 g	2.25 e	110.8 g
CV (%)	14.61	17.50	2.30

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Relationship between biological efficiency and age of fruiting body: Highly significant correlation between biological efficiency (BE) and age of fruiting body was observed. The relationship showed a quadratic equation as $y = -1.2432x^2 + 20.42x - 37.575$ ($R^2 = 0.8804^{**}$). The majority of total variation in BE of the mushroom can be explained by this equation. The R^2 value indicated that 88.04% of BE was attributed to age of fruiting body (Fig. 2). The response curve and the equation showed that BE was maximum at 6 day old fruiting body and it decreased at the rate of 1.2432% for per day change of age.

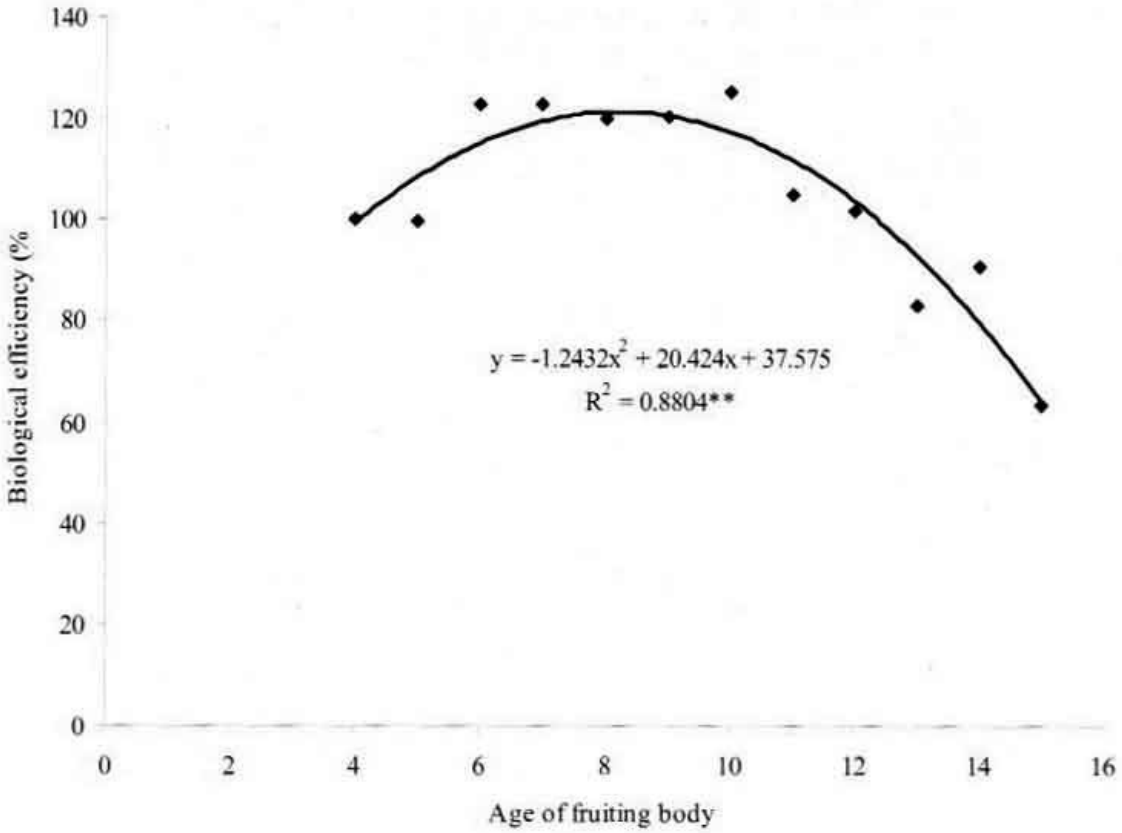

Fig. 2. Functional relationship between age of fruiting body and biological efficiency of milky mushroom.

In our study, the results reveal that mushroom should be harvested at 6th day after pinhead formation for good yield. Krishnamoorthy and Amutha (2007) also suggested to harvest milky mushroom in 6-8 days after pinhead formation.

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Mycelial Growth at Different Temperature and pH and Yield on Different Substrates of Oyster Mushroom var. White Snow

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Abstract

To study the effect of different temperature and pH on mycelial growth and substrates on yield of oyster mushroom var. white snow, six different temperatures- 10°C, 15°C, 20°C, 25°C, 30°C and 35°C, ten different pH levels- 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, 8.5 and 9.0 and eight different substrates- sawdust, coir pith, lentil straw, chickpea straw, lentil + chickpea straw, wheat straw, rice straw (autoclaved) and rice straw (hot water treated) were tested. The highest mycelium growth rate was observed at 20°C temperature and 6.5 pH level and the highest yield (242.8 g/ 500g substrate) obtained from hot water treated rice straw.

Key words: Temperature, pH, substrate, number of fruiting body, yield

INTRODUCTION

Cultivation of oyster mushroom is becoming popular throughout the world because of its ability to grow at a wide range of temperatures and to utilize various lignocelluloses (Baysal *et al.*, 2003). White snow variety of this mushroom is the new addition in the list of mushroom species of National Mushroom Development and Extension Centre, Savar, Dhaka, Bangladesh. It is very beautiful to look at as its color is milky white. Its taste and flavor are very good. The growth requirement of the mushroom is unknown and the suitable substrate is yet to be identified in Bangladesh. Therefore, the present piece of work was undertaken to study the mycelial growth in different temperature and pH for yield and yield attributes on different substrates of the mushroom.

MATERIALS AND METHODS

The present investigation was conducted at National Mushroom Development and Extension Centre, Savar, Dhaka from September to November 2010. Potato dextrose agar (PDA) medium was in use to evaluate the suitable temperature and pH for mycelium growth of oyster mushroom var. white snow. The culture media was prepared according to Moonmoon *et al.* (2008). For suitable temperature determination, the composition was adjusted at pH value 6.5 before autoclave. After cooling, the petri plates were inoculated with the inoculum of white snow mushroom. The inoculated plates were incubated at 10°C, 15°C, 20°C, 25°C, 30°C and 35°C temperatures. For pH study, ten different pH levels such as 4.5, 5.0, 5.5, 6.0, 6.5, 7.0, 7.5, 8.0, 8.5 and 9.0 of the media were tested for

getting the best for this particular mushroom. The pH levels were adjusted by adding 1N HCl or NaOH before autoclaving.

Eight different agricultural wastes such as sawdust, coir pith, lentil straw, chickpea straw, lentil + chickpea straw, wheat straw, rice straw (autoclaved) and rice straw (hot water treated) were tested as substrates. Spawn packets of all the substrates except the rice straw (hot water treated) were prepared, inoculated and incubated following the procedure mentioned by Sarker *et al.* (2007). The hot water treated rice straw was prepared following Shelly *et al.* (2010). All the spawn packets were of 500 g.

After completion of mycelium running, spawn packets were opened by D shaped cut on the shoulder and removed the pp sheet. The relative humidity and temperature of the culture house were maintained at 80-90% and 20-25^{0}C respectively by spraying water.

The experiment was laid out following completely randomized design (CRD) with four replications. Data on mycelial growth rate, days to completion of mycelium running, days required from opening to first harvest, length and diameter of pileus, numbers of fruiting body and yield were recorded and analyzed following standard methods using MSTAT-C computer program. Means were computed following DMRT using the same computer program.

RESULTS AND DISCUSSION

Effect of temperature: Highly significant difference was observed in mycelium growth rate at different temperatures (Table 1). The highest mycelium growth rate (0.35cm/ day) was observed in 20^{0}C which was statistically similar to that of 25^{0}C and significantly higher than 15 and 30^{0}C. No mycelium growth was observed in 10^{0}C and 35^{0}C temperature. Similar trend of mycelium growth was observed by Khandaker *et al.* (2009) and Imtiaz *el al.* (2008) in *Grifola frondosa* and *Agrocybe cylindracea*. The days to completion of mycelium running (DCMR) in different temperature were inversely proportional to the growth rate (Table 1). The lowest DCMR was observed in 20^{0}C which was significantly lower to all the temperature.

Table 1. Mycelium growth of oyster mushroom var. white snow at different temperature

Temperature (^{0}C)	Mycelium growth rate (cm/day)	Days to completion of mycelium running
10	-	-
15	0.27 bc	7.25 a
20	0.36 a	5.25 b
25	0.32 ab	7.00 a
30	0.26 c	8.00 a
35	-	-
C.V %	3.56	9.85

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Effect of pH: The highest mycelium growth rate (0.44 cm/ day) was observed in pH 6.5 which was significantly higher than all the pH levels (Table 2). The second highest mycelium growth rate (0.31cm/ day) was observed in pH 6.0 and 7.0 which was statistically similar to pH 7.5 (0.30 cm/ day). The lowest mycelium growth rate (0.11 cm/ day) was observed in pH 4.5 and 9.0. Similar observation was reported by Ibekwe *et al.* (2008) in case *Pleurotus ostreatus*. The days to completion of mycelium running were inversely proportional to the growth rate (Table 2).

Table 2. Mycelial growth of oyster mushroom var. white snow at different pH

pH	Mycelium growth rate (cm/day)	Days to completion of mycelium running
4.5	0.11 c	10.50 a
5.0	0.12 c	9.75 a
5.5	0.15 c	8.50 b
6.0	0.31 b	5.50 d
6.5	0.44 a	5.50 d
7.0	0.31 b	6.50 c
7.5	0.30 b	6.50 c
8.0	0.26 b	8.75 b
8.5	0.14 c	10.00 a
9.0	0.11 c	10.25 a
C.V (%)	5.37	7.15

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Effect of substrates: Days to completion of mycelium running (DCMR) ranged from 13.75 to 32.75 in different substrates and differed significantly (Table 3). The minimum days required for completion of mycelium running (13.75) was observed in hot water treated rice straw. Among the autoclaved substrates, the minimum days to completion of mycelium running (26.00) was observed in sawdust which was significantly lower than all other substrates. The highest days required for completion of mycelium running (32.75 cm) were observed in lentil + chickpea straw substrate.

The minimum days (4.75) required from opening to first harvest (DROFH) was observed in rice straw (hot water treated) substrate which was significantly lower than other substrates. Significantly higher DROFH was observed in rice straw (autoclaved) (8.75), lentil + chickpea straw (8.50) and chickpea straw (8.25) substrates.

The size of fruiting body was influenced by different substrates (Table 4). The highest length of pelius (7.45 cm) was observed in hot water treated rice straw which was statistically similar to autoclaved rice straw. The lowest length of pileus (3.92 cm) was observed in chickpea substrate. The diameter of pileus ranged from 0.60 to 1.20 cm. The highest diameter of pileus (1.20 cm) was observed in hot water treated rice straw which was statistically identical to sawdust. The lowest diameter of pileus (0.60 cm) was observed in the combination of lentil + chickpea straw substrate.

Table 3. Effect of different substrates on growth of oyster mushroom var. white snow

Substrates	Days to completion of mycelium running	Days required from opening to first harvest
Sawdust	26.00 c	5.75 d
Coir pith	29.25 b	7.25 c
Lentil straw	30.25 ab	7.50 bc
chickpea straw	31.50 ab	8.25 ab
Lentil + chickpea straw	32.75 a	8.50 a
Wheat straw	31.00 ab	7.250 c
Rice straw (autoclaved)	32.50 a	8.75 a
Rice Straw (hot water treated)	13.75 d	4.75 e
C.V %	5.53	7.18

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

The highest number of fruiting body (26.00) was found in sawdust substrate which is statistically similar to hot water treated rice straw. On the other hand, the lowest number of fruiting body (15.00) was obtained from autoclaved rice straw substrate. The highest yield (242.8 g/packet) was observed in hot water treated rice straw substrate which was significantly higher than other substrates. The lowest yield (125.50 g/packet) was obtained from coir pith substrate which was statistically similar to all other substrates except rice straw (hot water treated) and saw dust substrates. This result was supported by (Rajapakshe, *et al.*, 2007).

Table 4. Effect of different substrates on yield attributes and yields of oyster mushroom var. white snow

Substrate	Length of pileus (cm)	Diameter of pileus (cm)	Number of fruiting body	Yield (g/packet)
Sawdust	5.75 b	1.05 ab	26.00 a	188.8 b
Coir pith	5.2 b	0.92 b	24.00 ab	125.5 c
Lentil straw	4.90 b	1.00 b	16.50 bc	130.0 c
Chickpea straw	3.92 c	0.67 cd	16.50 bc	136.0 c
Lentil + Chickpea straw	4.75 bc	0.60 d	16.25 bc	138.8 c
Wheat straw	5.50 b	0.85 bc	16.00 bc	141.0 c
Rice straw (autoclaved)	6.95 a	0.90 b	15.00 c	138.8 c
Rice Straw (hot water treated)	7.4 a	1.20 a	22.00 abc	242.8 a
C.V %	11.19	14.61	26.94	6.18

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

The present results revealed that the highest mycelium growth rate was recorded at 20^0C with 6.5 pH. On the other hand, the minimum days required for mycelium running and the maximum yield (242.8 g/packet) obtained from hot water treated rice straw in oyster mushroom var. white snow.

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Effect of Different Plant Growth Regulators on the Yield and Yield attributes of White Button Mushroom

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Abstract

Two plant growth regulators, Gibberellic acid (GA_3) and Naphthalene acetic acid (NAA) were used at 20, 50, 100 and 200 ppm concentration to find out their effect on growth, yield and yield attributes of button mushroom. The lowest days required from casing to primordia initiation was 13.6 at 100 ppm GA_3. The higher number of fruiting body was recorded in NAA 50 ppm (70.6), GA_3 200 ppm (72.2) and NAA 100 ppm (67.8). The highest yield was recorded in NAA 100 ppm (348.62g/ 3kg of compost) followed by GA_3 50 ppm (336.52g/ 3kg of compost), NAA 50 ppm (336.09 GA_3 50 ppm (336.52g/ 3kg of compost) and GA_3 100 ppm (313.55g/ 3kg of compost).

Key words: *Agaricus bisporus,* NAA, GA_3, yield.

INTRODUCTION

White button mushroom contributes about 40% of the world mushroom production. The mushroom is popular as well as tasty. But the yield of this mushroom is very low as compared to other mushrooms like oyster, milky, straw etc. in Bangladesh condition. To increase yield of the mushroom a lot of research works is completed on substrates, supplements and post compost supplements. But very limited information is available on plant growth regulators. Plant growth regulators (PGRs) are organic compounds, other than nutrients, that modify plant physiological processes and act inside the plant cells to stimulate or inhibit specific enzymes or enzyme systems. These are normally active in low concentrations on different crops and help regulate plant metabolism. Mukhopadhyay *et al.* (2005) reported that the PGRs *viz.* IAA, GA3, KIN at different concentrations increase the biomass production of *Pleurotus sajor- caju* by 15-26%. The PGRs also increase the protein content of mycelia of the mushroom. Dey (1996) observed 72.33% increased production of *Pleurotus ostreatus* over control using 15ppm GA_3 in PDA media. Pani (2011) reported that biological efficiency was increased 82.2% in case of *Calocybe indica.* But almost no report is available in button mushroom. Plant growth

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regulators at lower concentration are ineffective and at higher concentration are detrimental (Pani, 2011) and all PGRs are not equally effective on all mushroom species. Thus, the present study was under taken to find out the effect of PGRs on the growth, yield and yield attributes of button mushroom and to determine the best possible concentration of the PGRs.

MATERIALS AND METHODS

The experiment was conducted in the Laboratory and Culture house of National Mushroom Development and Extension Centre, Savar, Dhaka during December 2006 to March 2008. Two plant growth regulators, Gibberellic acid (GA_3) and Naphthalene acetic acid (NAA) were used at the concentrations of 20 ppm, 50 ppm, 100 ppm and 200 ppm. The growth regulators were of Wako Pure Chemical Industry Ltd, Japan. The experiment was laid out following completely randomized design with 5 replications.

Preparation and use of growth regulator: To prepare 20 ppm of NAA and GA_3 solution, 20 mg of NAA and GA_3 powder were taken in two separate volumetric flasks and dissolved in 1 (N) Sodium Hydroxide and Ethyl alcohol respectively. The solvent were added as per requirement to dissolve the solute. Then the volume was made up to 1 L by adding distilled water followed by frequent stirring and 20 ppm solution was prepared. Similarly 50, 100 and 200 ppm solution of NAA and GA_3 were prepared by dissolving 50, 100 and 200 mg of respective powder. The solutions were sprayed during full mycelial growth (before and after casing), primordial initiation and fruiting body development stage. No watering was done after spraying the PGR on the same day.

Preparation of compost: Paddy straw was used as the main substrate and the compost was prepared by long method of composting (LMC) using rice straw (300 kg), wheat bran (30 kg), gypsum (15 kg), Calcium carbonate (10 kg), Urea (9 kg), Triple super phosphate (6 kg), Muriate of potash (3 kg), Furadan (250 g) and Bavistin (150 g). The ready compost was deep brown in colour, free from bad smell and had 65-67% moisture.

Spawning and incubation: Three kilograms of compost was mixed with 75 g of mother culture of *Agaricus bisporus* and poured in a polypropylene bag. The open top of the bags were covered with news papers and incubated in the incubation room at $24^0 \pm 2^0$C temperature for 20 days. The cover paper was wetted thrice a day to maintain the moisture level 65-67%.

Casing: After completion of mycelium run, the news paper sheet was removed and the surface of the compost was uniformly layered with 3.5- 4.0 cm casing materials. Before use, the casing materials were steam sterilized for 6 hour. The packets were incubated in the same incubation room at $24^o \pm 2^o$C temperature for 10 days.

Cropping and harvesting: Case run was considered complete when mycelia appeared in the valleys of casing layer. After case run, the spawn packets were transferred to a semi

controlled culture house where the temperature was decreased and relative humidity was increased by spraying water. The CO_2 concentration of the culture house was lowered by opening the windows for 10-15 minutes at 2-3 hours intervals. This change in environmental parameters induced pinhead formation within 3-4 days and developed into solid button sized mushrooms in another 3-4 days.

Mushrooms were harvested before the cap of the fruiting body showed any detachment from the stipe. Data were collected on days required to primordia initiation, number of fruiting body, biological efficiency (BE), yield and benefit cost ratio. The BE was estimated by the following formula-

BE (%) = Total biological yield (g) X 100/Total compost used (g).

Data were statistically analyzed and means were computed following Duncan's Multiple Test (DMRT).

RESULTS AND DISCUSSION

Days required from casing to primordia initiation: Days required from casing to primordia initiation (DRCPI) ranged from 13.6 to 15.2 days at 20-200 ppm level of giberellic acid (GA_3) and from 14.8 to 15.4 days at the same level of naphthalene acetic acid (NAA) (Table 1). The lowest DRCPI was recorded at 100 pm level of GA_3 which was significantly lower than all the treatment. The highest DRCPI (15.4) was recorded in NAA-200 ppm level which was statistically similar to all the treatments except 100 ppm level of GA_3.

Number fruiting body (NFB): The effect of plant growth regulators (GA_3 and NAA) on the number of fruiting body was significant among the treatments. Maximum number of fruiting body was recorded in NAA at 50 ppm which was significantly higher as compared to all other treatment except GA_3 at 200ppm and NAA at 100 ppm level (Table 1). Application of NAA in high concentration increased fruiting body formation by increasing cell division and enlargement. The lowest NFB was found in GA_3 at 20 ppm level which was significantly lower than GA_3 at 200 ppm and NAA at 50 and 100 ppm level.

Yield: The highest yield (348.62g/ 3kg of compost) was recorded in NAA at 100 ppm level which was statistically similar to NAA at 50 ppm and GA_3 at 50 and 100 ppm levels. The lowest yield (218.58 g/3kg compost) was recorded in NAA at 200 ppm level (Table 2). The result was supported by the statement of Charles (1986) who worked to find out the effect of growth regulators, cycocel and NAA on the yield and size of the commercial mushroom, *Agaricus bisporus* and noticed an increase in mean mushroom size without a reduction in yield. Despande and Temhane (1982) differed with this result as spraying with 200 mg/l IAA and NAA increased the yield of *Volvariella volvacea* by 10.6 and 8.0% respectively.

Table 1. Effect of different plant growth regulators on different yield contributing characters

Plant growth regulators	Concentration (ppm)	Days from casing to primordia initiation	Number of total fruiting body
Gibberellic acid (GA_3)	20	15.2 a	51.8 c
	50	14.8 a	60.4 bc
	100	13.6 b	60.8 bc
	200	15.0 a	70.2 a
Naphthalene Acetic Acid (NAA)	20	15.2 a	61.2 bc
	50	15.0 a	70.6 a
	100	14.8 a	67.8 ab
	200	15.4 a	52.0 c
Control	0	15.2 a	59.4 bc
CV (%)		4.87	10.57

Means within the column under a parameter having a common letter do not differ significantly (P=0.05).

Benefit cost Ratio (BCR): The highest BCR value 6.06 was estimated at NAA 100 ppm level of treatment, which was followed by NAA 50, GA_3 50, GA_3 20, NAA 20 and GA_3 100 ppm level. The lowest BCR (3.75) was estimated at NAA 200 ppm level which was followed by GA_3 200 ppm level and control (4.79) (Table 2).

Biological efficiency: Biological efficiency (BE) increased with the increase of the levels of the both GA_3 and NAA up to a certain level and then decreased (Fig. 1). The highest BE (12.91%) was observed in 100 ppm level of NAA. In case of GA_3, the highest BE (12.23%) was recorded in 50 ppm level and then decreased. From the study it is clear that both the PGR have a positive effect on BE of white button mushroom but have negative effect at higher concentration.

Relationship between levels of plant growth regulators and number of total fruiting body: The relationship of the levels of plant growth regulators (GA_3 and NAA) and the number of fruiting body was linear and quadratic respectively and could be expressed by the equations $y = 0.0699x^2 + 55.348$ ($R^2 = 0.7286$*) and $y = -0.0014x^2 + 0.2415x + 59.014$ ($R^2 = 0.9272$**). In case of GA_3, the regression equation ($y = 0.0699x + 55.348$, $R^2 = 0.7286$*) stated that the number of fruiting body increased gradually at the rate of 0.0699 per unit change of the levels of GA_3. The R^2 value indicated that 72.86% fruiting body was attributed to GA_3. In case of NAA, it is clear that the number of fruiting body of white button mushroom increased gradually with increased levels of NAA up to 50 ppm and started to decrease thereafter (Fig. 2).

Table 2. Effect of different plant growth regulators and their levels on the biological and economic yield and benefit cost ratio of white button mushroom

Plant growth regulators	Concentration (ppm)	Yield (g/3kg compost)	Benefit cost ratio
Gibberellic acid (GA_3)	20	306.07 bcd	5.36
	50	336.52 ab	5.65
	100	313.55 abc	5.01
	200	291.85 cd	4.27
Naphthalene Acetic Acid (NAA)	20	287.00 cd	5.06
	50	336.09 ab	5.92
	100	348.62 a	6.06
	200	218.58 f	3.75
Control	0	247.85 ef	4.79
CV (%)		8.95	

Means within the column under a parameter having a common letter do not differ significantly (P=0.05).

Relationship between levels of plant growth regulators and economic yield: There was significant correlation between level of NAA and yield of white button mushroom (Fig. 1). The relationship between level of NAA and yield could be expressed by quadratic equation as $y = -0.0119x^2 + 2.2243x + 248.86$ ($R^2 = 0.9959^{**}$) where y = economic yield and x = level of NAA. The R^2 value indicated that 99.59% of yield of white button mushroom was attributed to the level of NAA and the yield was decreased at the rate of 0.0119 g/bag for per unit change of level of NAA (x). No significant relationship was observed between levels of GA_3 and Economic yield (Fig. 3).

Spraying of NAA and GA_3 at 50 to 100 ppm during cropping period may be recommended for higher yield of white button mushroom.

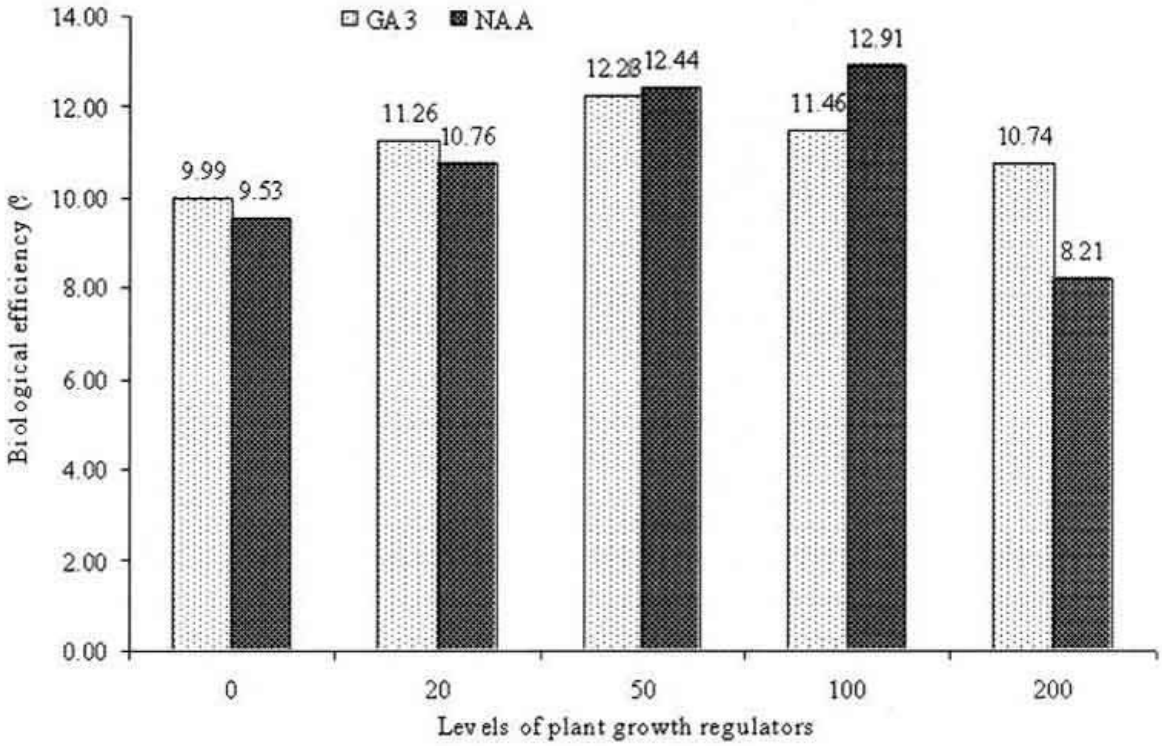

Fig. 1. Effect of different levels of plant growth regulators on the biological efficiency of white button mushroom.

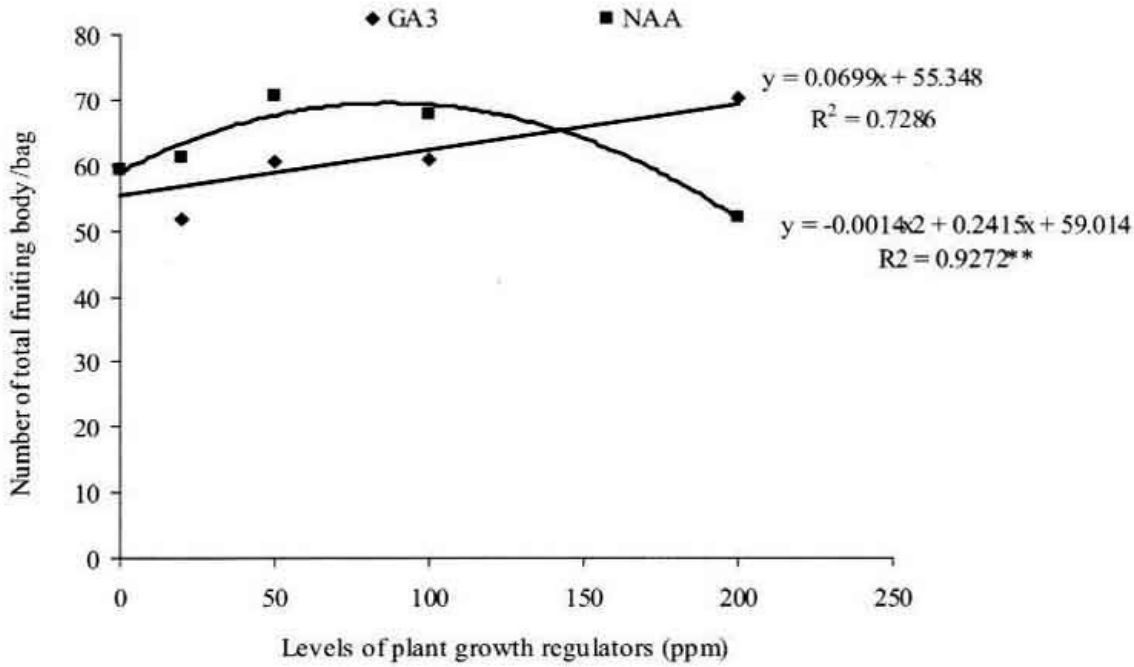

Fig. 2. Functional relationship between levels of plant growth regulators and number of total fruiting body of white button mushroom.

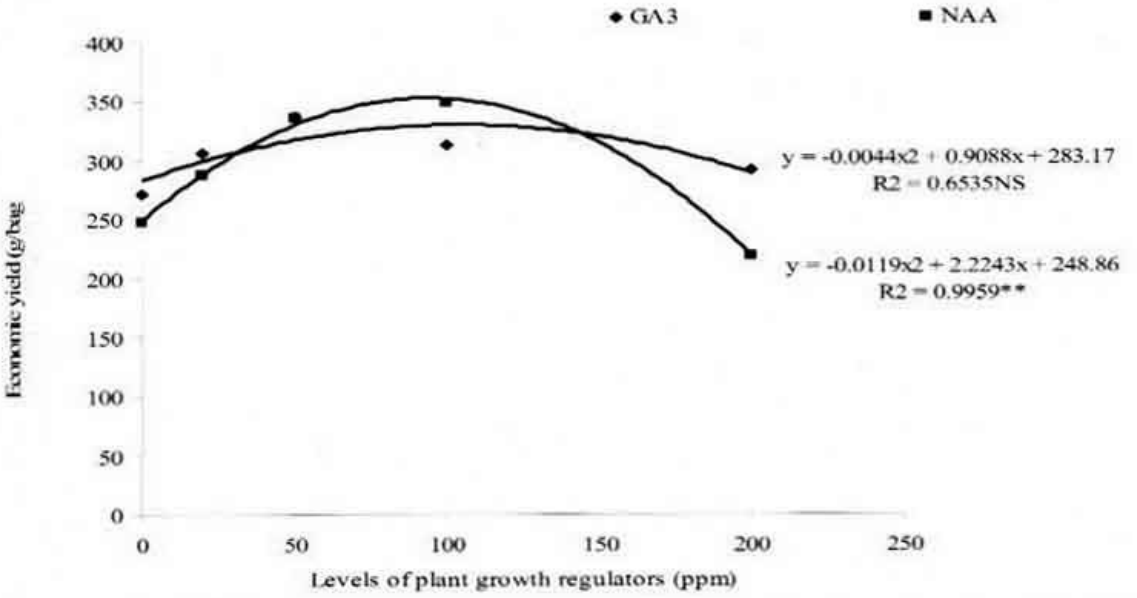

Fig. 3. Relationship between levels of plant growth regulators and economic yield of white button mushroom.

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Performance of different strains of *Pleurotus cystidiosus* under Bangladesh condition

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Abstract

Performance of different strains of *Pleurotus cystidiosus* (Pcys) were studied in National Mushroom Development and Extension Centre, Savar, Dhaka, Bangladesh. Significant variation was observed in the growth, yield and yield contributing characters of these strains. The highest mycelial growth (0.58 cm/day) was observed in Pcys-4 which was followed by Pcys-5, Pcys-2 and Pcys-1. The lowest mycelial growth (0.22 cm/day) was observed in Pcys-3. The highest biological and economic yield, 196.3 and 189.0 g/packet respectively were obtained from Pcys-1, which followed by Pcys-2, Pcys-4 and Pcys-5. The lowest biological and economic yields were observed in Pcys-6. The number of effective fruiting bodies was the highest (37.25) in pcys-1, while, the weight of individual fruiting body was the highest (26.88 g) in Pcys-6. The highest length of stipe (4.95cm) and thickness of pileus (1.35cm) were observed in Pcys-1. The strain also showed the highest biological efficiency (BE) among the strains.

Key words: *Pleurotus cystidiosus*, Growth, yield attributes, yield.

INTRODUCTION

Production of oyster mushroom has increased greatly throughout the world during the last few decades and its popularity is increasing day by day due to its simple cultivation techniques and high yield potential (Banik and Nandi, 2004). A number of species of the mushroom is being cultivated throughout the world, such as the tree oyster (*Pleurotus ostreatus*), the gray oyster (*Pleurotus sajor-caju*), the abalone oyster (*Pleurotus cystidiosus*), the white oyster (*Pleurotus florida*), the golden oyster (*Pleurotus citrinopileatus*), the pink oyster (*Pleurotus salmoneostramineus*) and the black oyster (*Pleurotus sapidus*). Among the species, the abalone mushroom (*Pleurotus cystidiosus*) is newly introduced in Bangladesh which was first described by Orson K. Miller in 1969 from a maple tree in Indiana, USA. The mushroom is gaining popularity in Bangladesh for its beautiful size, shape and colour. A number of strains of the mushroom are available in Bangladesh, though all of them are not performing well round the year. The purpose of this study was to investigate the performance of different strains of *Pleurotus cystidiosus* in order to identify the best strain that can be highly productive and suitable for the cultivation in the winter in Bangladesh.

MATERIALS AND METHODS

The experiment was conducted in the National Mushroom Development and Extension centre (NAMDEC), Sobhanbag, Savar, Dhaka during November 2010 to February 2011. Six different strains of *Pleurotus cystidiosus* (Pcys) available in NAMDEC were selected for the experiment. The strains were designated as Pcys-1, Pcys-2, Pcys-3, Pcys-4, Pcys-5 and Pcys-6.

Preparation of pure culture: Pure cultures of selected strains were grown on potato dextrose agar (PDA) medium. The medium was poured into test-tube. The medium in test-tube was autoclaved at 120^0C and 1 kg/cm^2 for 20 minutes. After sterilization and solidification, the test-tubes were inoculated with the inocula of the strains. All operations were done under sterile condition in a clean bench. The inoculated test-tube were incubated in a growth chamber at 20 ± 2^0C and 60-70% relative humidity (RH). After completion of the mycelium run, this culture was used for inoculation of mother culture.

Preparation of mother culture: Mother culture was prepared on maize grain. The grain was put into water for an hour, cooked for 40 minutes and washed in flowing water. The excess water of the grain drained off. The grain was supplemented with 0.2% $CaCO_3$ and was mixed manually. The moisture could be of 60-65%. Polypropylene bags of 7″ x 10″ size were filled with 200 g maize and their mouths were plugged by inserting water absorbing cotton with the help of plastic neck. The bags were autoclaved at 121^0C and 1kg/cm^2 pressure for 2 hours. After autoclaving and cooling, the bags were inoculated with the pure culture of *Pleurotus cystidiosus* mushroom strains at the rate of 1 cm^2 culture slant per packet. After inoculation, the packets were incubated in the laboratory at about 20 ± 2^0C temperature.

Preparation of spawn packets: Sawdust was thoroughly mixed with nutrient materials, wheat bran at the ratio of 2:1, water was added to make the moisture content 60% and $CaCO_3$ was added at the rate of 0.2% of the total mixture. Polypropylene bags of 7″ x 10″ size were filled with 500 g mixture of substrate and their mouths were plugged by inserting water absorbent cotton with the help of plastic neck. The bags were autoclaved at 121^0C and 1kg/cm^2 pressure for 2 hours. After autoclaving and cooling, the bags were inoculated with the mother culture of *Pleurotus cystidiosus* mushroom strains at the rate of 1 teaspoonful per packet. After inoculation, the packets were incubated in the laboratory at about 20 ± 2^0C temperature.

After completion of mycelium running, spawn packets were opened by 'D' shape cut on both the shoulder of the packets and transferred to culture house at 14-20^0C and 60-70% relative humidity.

Data collection and analysis: The experiment was laid out in a completely randomized design (CRD) with four replications. Yield and yield parameters were taken on the basis of three flashes except in pcys-6 where only one flash was obtained. Data on mycelial growth rate, days required from stimulation to primordia initiation, number of primordia, number of fruiting body, number of effective fruiting body, length of stipe, diameter of

stipe, diameter of pileus, thickness of pileus, biological yield (g/packet) and economic yield (g/packet) were recorded. Data were analyzed followed by Gomez and Gomez (1984) using MSTAT-C computer program. Means were computed following Duncan's multiple Ranges Test (DMRT).

RESULTS AND DISCUSSION

Mycelial growth: Mycelial growth rate varied from 0.22 cm to 0.58 cm/day (Table 1). The highest growth rate (0.58 cm/day) was observed in Pcys-4 which was followed by Pcys-5, Pcys-2 and Pcys-1. The lowest mycelium growth rate (0.22 cm/day) was observed in Pcys-3.

Days required from stimulation to primordia initiation: Days required from stimulation to primordial initiation (DRSPI) ranged from 3.50 to 12.25 (Table 1). The highest DRSPI was found in pcys-6 (12.25 cm/day) and the lowest DRSPI was found in pcys-4 (3.50 days) proceeded by pcys-2 (4.50 days). The result is almost similar with Patra and Pani (1995) who reported that oyster mushroom took 4 to 8 days for initiation of primordia. It was also similar with the findings of Shelly *et al.* (2009) who reported that *Pleurotus cystidiosus* took 7 to 8 days for initiation of primordia.

Table 1. Mycelial growth and primordia initiation in different strains of *Pleurotus cystidiosus*

Strains	Mycelial growth (cm/day)	Primordial initiation (days)
Pcys-1	0.25 cd	6.50 b
Pcys-2	0.28 c	4.50 cd
Pcys-3	0.22 d	6.00 b
Pcys-4	0.58 a	3.50 d
Pcys-5	0.54 b	5.25 bc
Pcys-6	0.23 d	12.25 a
CV (%)	7.74 %	16.85

In a column, the same letters are not significantly different by Duncan's multiple range test at 5% level.

Number of primordia: The number of primordia obtained from 3 flushes (except in pcys-6) in different treatment differed significantly (Table 2). The highest number of primordia was observed in pcys-2 (85.25) which was statistically similar to Pcys-1 (77.75) and the lowest number of primordia was observed in pcys-6 (1.50). The result was found similar to Amin *et al.* (2007) who observed that the highest number of primordial/packet from 3 flushes was 81 in case of oyster mushroom.

Number of fruiting body: The number of fruiting bodies obtained from 3 flushes (except in pcys-6) in different treatments varied significantly and ranged from 1.50 to 45.50

(Table 2). The highest numbers of fruiting bodies were found in pcys-2 (45.50) which was statistically similar to Pcys-1 (43.00) and the lowest numbers of fruiting bodies were found in pcys-6 (1.50).

Number of effective fruiting bodies: The number of effective fruiting bodies obtained from 3 flushes (except in pcys-6) in different treatments varied significantly (Table 2). The highest numbers of effective fruiting bodies were found in pcys-1 (37.25) followed by pcys-2 (35.50) and the lowest numbers of fruiting bodies were found in pcys-6 (1.50).

Weight of individual fruiting bodies: Weight of individual fruiting body in different treatments ranged from 4.78 g to 26.88g without any significant differences (Table 2). The weight of individual fruiting body was highest in pcys-6 (26.88g) and lowest in pcys-3 (4.78g) followed by pcys-4 (4.92g), pcys-2 (4.95g), pcys-1 (5.43g) and pcys-5 (6.34g).

Length of stipe: The length of stipe ranged from 1.33 cm to 4.95 cm (Table 3). The highest length of stipe was found in pcys-1 (4.95 cm) and the lowest was found in pcys-3 (1.33 cm). More or less similar result was obtained by Alam *et al.* (2007) in case of oyster mushroom. It's also similar that Shelly *et al.* (2009) in case of *Pleurotus cystidiosus.*

Table 2. Yield attributes in different strains of *Pleurotus cystidiosus*

Mushroom strains	Number of primordia	Number of fruiting body	Number of effective fruiting body	Weight of individual fruiting body (g)
pcys-1	77.75 a	43.00 a	37.25 a	5.43 b
pcys-2	85.25 a	45.50 a	35.50 a	4.95 b
Pcys-3	53.75 b	30.50 b	23.25 bc	4.78 b
pcys-4	62.00 b	35.25 b	25.75 b	4.92 b
pcys-5	60.25 b	29.75 b	19.00 c	6.34 b
Pcys-6	1.50 c	1.50 c	1.50 d	26.88 a
CV (%)	16.59	18.96	17.96	38.59

In a column, the same letters are not significantly different by Duncan's multiple range test at 5% level.

Diameter of stipe: The diameter of stipe differed significantly and ranged from 0.95 cm to 1.43 cm (Table 3). The highest diameter of stipe was found in pcys-6 (1.43 cm) followed by pcys-1 (1.35 cm), pcys-4 (1.18 cm) and the lowest diameter was found in pcys-5 (0.95 cm).

Diameter of pileus: The diameter of pileus differed significantly and ranged from 5.28 cm to 9.75 cm (Table 3). The highest diameter of pileus was found in pcys-6 (9.75 cm) and the lowest diameter of pileus was found in pcys-3 (5.28 cm).

Thickness of pileus: The thickness of pileus of different strains differed significantly and ranged from 0.49 cm to 1.35 cm (Table 3). The highest thickness was found in pcys-1 (1.35 cm) and the lowest thickness was found in pcys-3 (0.49 cm).

Table 3. Size of fruiting body in different strains of *Pleurotus cystidiosus*

Mushroom strains	Length of stipe (cm)	Diameter of stipe (cm)	Diameter of pileus (cm)	Thickness of pileus (cm)
pcys-1	4.95 a	1.35 ab	7.58 b	1.35 a
pcys-2	3.62 b	1.10 bc	6.40 c	0.97 b
Pcys-3	3.33 b	1.06 c	5.28 d	0.49 c
pcys-4	3.38 b	1.18 abc	8.30 b	0.65 c
pcys-5	1.50 c	0.95 c	8.03 b	0.70 c
Pcys-6	1.33 c	1.43 a	9.75 a	1.25 a
CV (%)	14.19	17.59	11.19	21.85

In a column, the same letters are not significantly different by Duncan's multiple range test at 5% level.

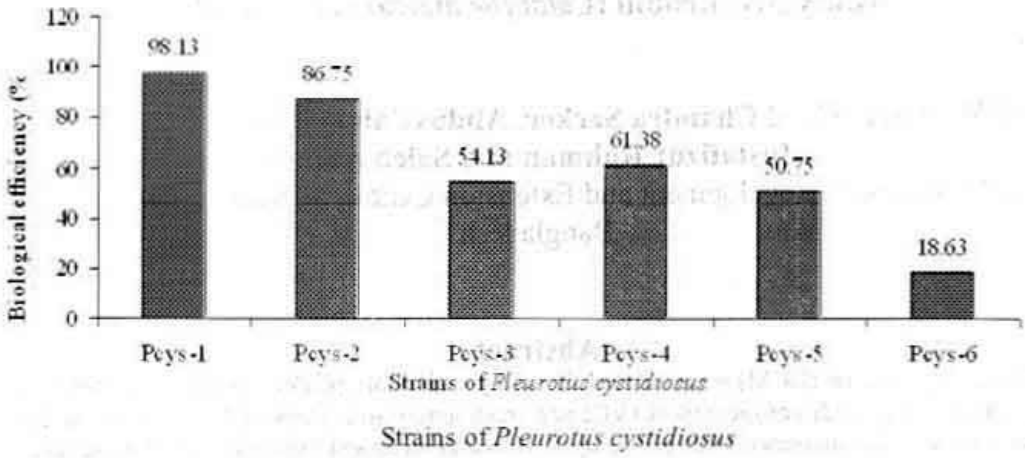
Yield: Significant variation was observed in biological yield at 5% level of significance. The biological yield from 3 flushes (except in pcys-6) ranged from 37.25 g to 196.3 g/packet (Table 4). The highest biological yield was found in Pcys-1 (196.3 g/packet) and the lowest biological yield was found in Pcys-6 (37.25 g/packet). Similar trend was observed in economic yield (Table 4).

Table 4. Biological and economic yield in different strains of *Pleurotus cystidiosus*

Mushroom Species	Biological yield (g/ packet)	Economic yield (g/ packet)
pcys-1	196.3 a	189.0 a
pcys-2	173.5 b	166.3 b
Pcys-3	108.3 cd	98.25 cd
pcys-4	122.8 c	116.8 c
pcys-5	101.5 d	95.00 d
Pcys-6	37.25 e	35.50 e
CV (%)	12.15	12.99

In a column, the same letters are not significantly different by Duncan's multiple range test at 5% level.

Biological efficiency: Remarkable variation was observed in biological efficiency of six strains of *Pleurotus cystidiosus* (Fig.1). The highest biological efficiency (98.13%) was obtained from Pcys-1 followed by Pcys-2, Pcys-4 and Pcys-3. The lowest biological efficiency (18.63%) was found in Pcys-6.



Strains of *Pleurotus cystidiosus*

Fig. 1. Biological efficiency of six cultivated strains of *Pleurotus cystidiosus*.

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Reuse of Casing Materials Amended with Vermicompost and Loamy Soil in Milky Mushroom (*Calocybe indica*) Cultivation

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Abstract

Used casing material (UCM) was recycled for milky mushroom (*Calocybe indica*) cultivation after amendment with vermicompost (VC) and fresh loamy soil. Days to first harvest (DFH) decreased with the increased rates of VC up to 10% and increased thereafter. The lowest DFH (17.50) was observed in 10% VC, fresh loamy soil (soil+sand, 3:1) (FLS) and when the UCM and FLS were mixed at 1:2 ratios. The highest number of fruiting body (14.5) was observed in FLS, UCM + FLS (1:1) and UCM + FLS (1:2). The highest yield (483.30g/ packet) was observed in UCM + FLS (1:1) followed by FLS (460.80 g/packet) and UCM + FLS (1:2) (441.50 g/packet). Maximum biological efficiency (BE) was achieved with UCM + FLS (1:1) followed by FLS and UCM + FLS (1:2).

Key words: Milky mushroom, casing materials, vermicompost, loamy soil, yield.

INTRODUCTION

Casing is a layer of materials that covers the top of mushroom bed after completion of spawn run. It enhances the transformation of vegetative phase of mushroom fungi to reproductive phase to initiate fruiting body. In western countries magnum moss and peat are used as the casing material. In Bangladesh loamy soil is used as casing material for milky mushroom cultivation. In urban area, collection of new soil and dumping of the used casing material is a laboursome and costly activity. So, it is very important to recycle the used casing materials. Generally, macro- and micro-nutrients are exhausted from used casing materials. To compensate the shortage of the nutrients, used casing materials may be amended with vermicompost and fresh loamy soil, which are good supplements to rejuvenate the used casing materials for recycling. The vermicompost supplement improves both nutritional and microbial status of casing materials (Nagratna and Mallesha, 2007). The vermicompost supplies macronutrients like nitrogen, phosphorous and potash and also micronutrients like iron, zinc, calcium, magnesium and copper (Kale, 1998). Use of vermicompost as an additive for formulation of casing materials increases yield of *Agaricus bisporus* (Garcia *et al.* 2005). Umamaheshwari and Vijayalakshmi (2004) found that utilization of earthworm casts as casing material helps in checking water loss by evaporation and the water holding capacity of the casts contributes to the increased yield. Dhar *et al.* (2003) reported that the use of different agricultural wastes including vermicompost in the casing material increased the nitrogen content and total mushroom

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yield of *A. bisporus*. The present experiment was conducted to find out the effect of amendment of used casing materials with vermicompost and fresh loamy soil on its rejuvenation to cultivate milky mushroom.

MATERIALS AND METHODS

The experiments were conducted at National Mushroom Development and Extension Centre, Sobhanbag, Savar, Dhaka during May 2010 to August 2010. The used casing materials (UCM) were loamy soil, prepared by mixing soil and sand at the ratio 3:1 before use in the previous crop. The UCM was amended with vermicompost (VC) or fresh loamy soil (soil + sand, 3:1) (FLS) at different rates to have following treatments: T_1= UCM without amendment, T_2= UCM amended with 5% VC, T_3= UCM amended with 10% VC, T_4= UCM amended with 15% VC, T_5= UCM amended with 20% VC, T_6= UCM amended with 50% VC, T_7= FLS, T_8= UCM amended with FLS (1:1) and T_9= UCM+FLS (1:2).

Mother culture, a seed material for mushroom cultivation, was prepared following standard procedure (Krishnamoorthy, 1981). For this purpose, apparently healthy and clean wheat grains were boiled with equal amount of water till grains become soft. They were not allowed to split open. The moisture content of boiled grains was allowed to leave as such for hours and for air dried so that water on surface is evaporated to obtain 50 to 55% moisture content. The grains were mixed with 0.5% calcium carbonate so that the pH of the grain is around 7.0 to 7.8 and they do not form clumps. This substrate was poured into 7" × 10" size polypropylene bags at 300 g/bag and sterilized in an autoclave for 1 hour at 120^{0}C. After cooling for 24 hours, the content was inoculated with fresh mycelial culture of *Calocybe indica* under aseptic condition. The containers were incubated at 26-28°C for 15-20 days for *Calocybe indica* to attain complete ramification over the substrate. Freshly prepared seed materials were used for cultivation of aforementioned mushroom.

Rice straw substrate was used for the cultivation of *Calocybe indica*. Rice straw was chopped into 6 to 10 cm in length. The substrate was treated with hot water at 60^{0}C for 60 minutes and allowed to drain off the excess water by hanging the straw bags for 20 hours. The hot water treated straw was spread on a clean concrete floor for air drying to remove excess moisture. Polypropylene bags (12″×18″) were filled with straw substrate and seeded with 5% *Calocybe indica* grain mother following method (Krishnamoorthy, 1981). The bags were incubated in a culture room 25 ± 5°C. After 18-22 days of incubation the substrate was completely colonized by the mycelium and the cover of the polypropylene bag was opened.

The amended used casing materials under different treatments were sterilized at 65^0 C for 4 hrs and used as casing material. Casing materials were covered over the mycelial growth on the substrate up to 4 cm thickness. Watering was done at regular interval to maintain moisture level at 60 to 70%. Pin head appeared at 14-17 days and developed into fruiting bodies. Matured fruiting bodies were harvested and data on days required from

casing to first harvest, length of stipe, diameter of stipe, diameter of pileus, thickness of pileus, number of fruiting body, yield and biological efficiency (BE) of the substrate were recorded. The BE was measured using the formula: BE= Fresh weight of mushroom X 100/ Dry weight of substrate.

The experiment was laid out in a completely randomized design with 4 replications. The data were statistically analyzed using MSTAT-C computer programme. Means were compared following Duncan's Multiple Range Test (DMRT) using the same computer programme.

RESULTS AND DISCUSSION

Days to first harvest: The lowest days to first harvest (DFH) (17.50) was recorded under the treatments T_3 (UCM + VC 10%), T_7 (only FLS) and T_9= (UCM+FLS, 1:2), which was statistically similar to T_8 (UCM + FLS, 1:1) but significantly lower to other treatments. The highest DFH was recorded from T_6 (UCM + VC, 50%), which was significantly different from all other treatments except T_1 (Only UCM). The DFH under T_2 (UCM + VC, 5%), T_4 (UCM + VC, 15%), T_5 (UCM + VC, 20%) and T_8 (UCM + FLS, 1:1) ranged from 19.50-21.50. However, their differences were not significant. Days to first harvest (DFH) decreased with the increase of vermicompost (VC) added to used casing material (UCM) up to 10% and increased thereafter. The result reveals that VC has positive effect on DFH up to a certain level and at higher dose it has negative effect on this parameter (Table 1).

Size of fruiting body: The length of stipe under different treatments ranged from 6.32 to 9.55 cm. The highest length of stipe was recorded from T_9 (9.55 cm) followed by T_7 (9.02 cm), T_3 (8.72 cm), T_4 (8.50 cm), T_5 (7.45 cm) and T_2 (7.02 cm). Their differences were significant. The lowest length of stipe was found in T_1 (6.32 cm), which was statistically similar to T_6 (6.40 cm) (Table 1). The highest diameter of stipe was found in T_5 (3.35 cm), which was statistically similar to T_4 and T_9 (3.25 cm). The lowest diameter of stipe was observed in T_6 (2.45 cm), which was statistically similar to T_2 (2.55cm) (Table 1).

The highest diameter of pileus was obtained from T_9 (7.12 cm) followed by T_2 (6.95 cm) and T_8 (6.92 cm). The lowest diameter of pileus was observed in T_1 (5.45 cm) followed by T_6 (5.65 cm) and, T_3 and T_4 (6.65). Their differences were significant. Thickness of pileus under treatments T_2, T_3, T_4, T_6, T_7, T_8 and T_9 were statistically similar and significantly higher to T_1 and T_6 (Table 1). The lowest thickness of pileus was recorded from T_6 (0.95 cm) followed by T_1 (1.15 cm).

Table 1. Effect of used casing materials amendment with vermicompost and fresh loamy soil on days to first harvest and size of fruiting structures of milky mushroom (*Calocybe indica*)

Treatment*	Days to first harvest	Length of stipe (cm)	Diameter of stipe (cm)	Diameter of pileus (cm)	Thickness of pileus (cm)
T_1= Only UCM	21.50 ab	6.32 g	2.95 c	5.45 g	1.15 b
T_2=UCM+VC 5%	20.50 b	7.02 f	2.55 de	6.95 ab	1.9 a
T_3=UCM+VC 10%	17.50 c	8.72 c	3.05 c	6.65 d	1.8 a
T_4=UCM+VC 15%	20.50 b	8.50 d	3.25 ab	6.65 d	2.05 a
T_5=UCM+VC 20%	20.50 b	7.45 e	3.35 a	6.75 cd	1.85 a
T_6=UCM+VC 50%	22.75 a	6.40 g	2.45 e	5.65 f	0.95 c
T_7= only FLS	17.50 c	9.02 b	2.65 d	6.10 e	2.05 a
T_8=UCM+FLS (1:1)	19.50 bc	8.52 d	3.05 bc	6.92 bc	2.05 a
T_9=UCM+FLS (1:2)	17.50 c	9.55 a	3.25 ab	7.12 a	2.05 a
CV (%)	6.80	2.72	4.38	1.87	7.28

In a column, means followed by a common letter are not significantly different at 5% level by DMRT. *UCM = Used casing material of loamy soil (soil+sand, 3:1), VC=Vermicompost, FLS = Fresh loamy soil (soil+sand, 3:1).

Number of fruiting body/packet: The maximum number of fruiting body was found in treatments T_6 and T_8 (14.5), which were statistically similar to T_5 and T_8 (Table 2). The lowest number of fruiting body (7.5) was recorded in T_1 and T_6 followed by T_4, T_2 and T_3. The number of fruiting body increased with the increases of VC upto 10%. The fresh casing materials performed better than amended used casing materials (Table 2).

Yield: The highest yield was obtained from T_8 (483.30g/ packet) followed by T_7 (460.80 g/ packet), T_9 (441.50) and T_3 (394.30 g/packet). Their differences were significant. The lowest yield was found in T_1 followed by T_6, T_2, T_5 and T_4 which yielded 200.80, 248.80, 333.00, 352.00 and 383.80 gram mushroom per packet (Table 2).

Biological efficiency: The higher biological efficiency was achieved with T_8, T_7 and T_9. Their effect on this parameter was statistically similar and significantly higher compared to other treatments. The lowest biological efficiency was recorded from T_1 followed by T_6. The effect of the treatments T_2, T_3, T_4 and T_5 on biological efficiency was statistically similar and significantly higher compared to T_1 and T_8 but lower compared to other three treatments (Fig. 1).

The results of the present experiment reveal that the efficiency of used casing materials was enhanced due to amendment with vermicomposts. Effectiveness of vermicompost increased with the increase of its level up to 10% and decreased thereafter. The vermicompost had negative effect at higher dose on the parameter. The results were in line with those of Tandon *et al.* (2006) who reported that vermicompost gave the lowest

yield. The low yield with higher vermicompost level could be attributed to its high salinity that results in yield reduction as reported in case of *Agarcus bisporus* (Kurtzman, 1995).

Table 2. Effect of amendment of used casing materials with vermicompost and fresh loamy soil on number of fruiting body and yield of milky mushroom (*Calocybe indica*)

Treatment	Number of fruiting body/packet	Yield (g/ packet)
T_1= Only UCM	7.50 d	200.80 h
T_2=UCM+VC (5%)	10.50 c	333.00 f
T_3=UCM+VC (10%)	11.50 bc	394.30 d
T_4=UCM+VC (15%)	9.75 c	383.8 d
T_5=UCM+VC (20%)	13.50 ab	352.00 e
T_6=UCM+VC (50%)	7.50 d	248.80 g
T_7= only FLS	14.50 a	460.80 b
T_8=UCM+FLS (1:1)	14.00 a	483.30 a
T_9=UCM+FLS (1:2)	14.50 a	441.50 c
CV (%)	12.30	2.95

In a column, means followed by a common letter are not significantly different at 5% level by DMRT. [UCM = Used casing material of loamy soil (soil+sand, 3:1), VC=Vermicompost, FLS = Fresh loamy soil (soil+sand, 3:1)].

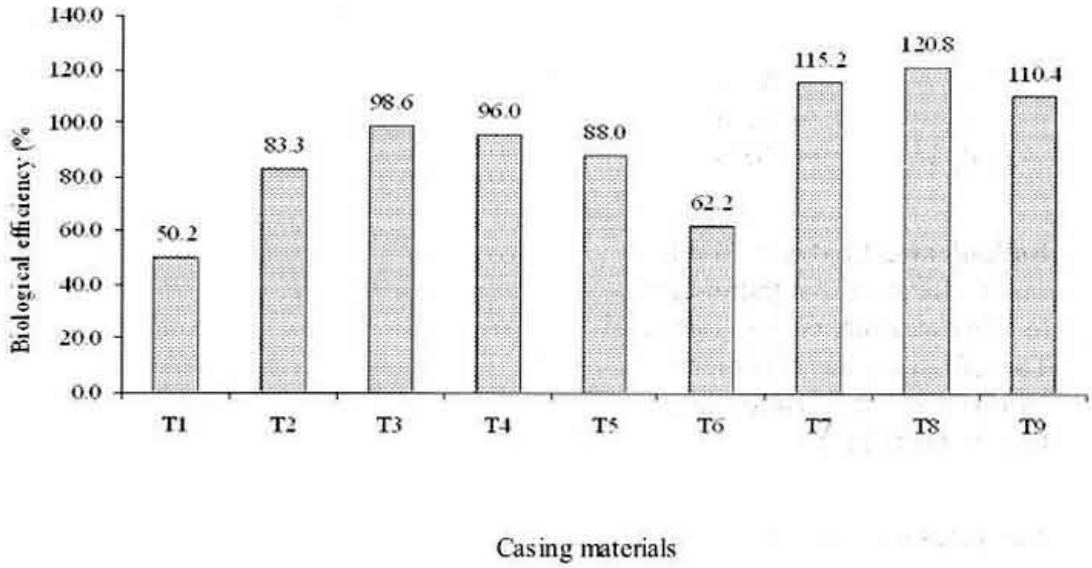

Fig. 1. Effect of casing material on biological efficiency of milky mushroom [T_1= Only UCM, T_2=UCM+VC (5%), T_3=UCM+VC (10%), T_4=UCM+VC (15%), T_5=UCM+VC (20%), T_6=UCM+VC (50%), T_7= only FLS, T_8=UCM+FLS (1:1) and T_9=UCM+FLS (1:2), UCM = Used casing material of loamy soil (soil+sand, 3:1), VC=Vermicompost, FLS = Fresh loamy soil (soil+sand, 3:1)].

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Effect of Different Supplements on Yield and Nutritional Status of Shaggy Mushroom

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Abstract

Sawdust was supplemented with wheat bran (WB), rice bran (RB), maize powder (MP), WB+RB (1:1), WB+MP (1:1) and RB+MP (1:1) to cultivate shaggy mushroom. The ratio of supplements to substrate was 1:2. Significant variation was observed in the number and size of fruiting body and yield of the mushroom. The higher number of fruiting body was observed in RB+WB (1:1) (9.0) and WB (7.75) supplement. The higher yield (170.0 g/ packet) was also recorded in RB+WB (1:1) supplement. Carbohydrate and protein content of shaggy mushroom was remarkably influenced by different supplements and the highest amount of carbohydrate (41.75%) and protein (29.75%) were observed in the mushrooms grown on MP and WB + RB (1:1) respectively. No remarkable difference was observed in fat, fiber and ash content of shaggy mushroom grown on sawdust supplemented with different supplements.

Key words: *Coprinus comatus*, sawdust, supplement, yield, nutritional quality.

INTRODUCTION

Shaggy mushroom (*Coprinus comatus*) is a very common, visually distinctive mushroom with a nice flavor. It is a quite popular mushroom. The young fruiting bodies first appear as white cylinders emerging from the ground and then the bell-shaped caps open out. Shaggy mushroom prefers the substrate like sawdust, rice straw etc. (Sarker, *et al.*, 2008). In order to increase the production of mushrooms, there is need to improve on cultivation and growth procedure. Growing mushrooms on simple substrate alone sometimes cannot provide enough nitrogen required for optimal growth of mushrooms. Supplements may be added to obtain higher yields. Many kinds of waste materials such as wheat bran, rice bran and maize powder are used for supplementation with the substrate at different level. These supplements also raise the nutritional status of the mushroom. An and Awan (1996) obtained the higher yield with the use of rice straw as the main substrate and rice bran as additives. The purpose of this study was to find out the most suitable supplement to sawdust substrate for shaggy mushroom cultivation and to determine the nutritional status of the mushroom influenced by the supplements.

MATERIALS AND METHODS

The experiment was conducted at the National Mushroom Development and Extension Centre, Savar, Dhaka during November 2010 to January 2011. The substrate (sawdust)

was supplemented with wheat bran (WB), rice bran (RB) and maize powder (MP) and their combinations, WB+RB (1:1), WB+MP (1:1) and RB+MP (1:1) at the ratio of 2:1. Water was added to make the moisture content 60%. Calcium carbonate ($CaCO_3$) was added at the rate of 0.2% of the total mixture. After mixing, the polypropylene bags of 18 x 25 cm size were filled with 500 g of substrates. The bags were plugged with absorbent cotton covering by brown paper with the help of a rubber band and autoclaved at 121^0C temperature and 1.5 kg/ cm^2 pressure for 2 hours. The autoclaved bags were allowed to cool for about 12 hours and inoculated with the inoculums of *Coprinus comatus* at the rate of 2 table spoon full per packet. The inoculated bags were incubated for 25 days at about 22^0C temperature. After completion of mycelial running, the bags were opened and casing soil was sprayed over the mycelium at the thickness of 2- 3 cm. The bags were transferred to culture house at cool temperature for fruit body formation.

The experiment was laid out following completely randomized design (CRD) with four replications. Data on number of fruiting body, yield and length and diameter of both stalk and pileus were recorded and analyzed using MSTATC computer program. Means were separated following DMRT using the same computer program.

Analytical methods of nutritional analysis: The nutrient value of fruit bodies was determined in terms of protein (Burties and Ashwood, 2006), fat (Folch *et al.*, 1957), carbohydrate (Raghuramalu *et al.*, 2003), fiber (Raghuramalu *et al.*, 2003) and total ash (Raghuramalu *et al.*, 2003).

RESULTS AND DISCUSSION

Number of fruiting body: Significant variation was observed in number of fruiting body of shaggy mushroom in different supplements to sawdust substrates. The highest number of fruiting body (9.0) was observed in rice bran (RB)+wheat bran (WB) (1:1) supplement, which was significantly higher to other supplements except WB. The lowest number of fruiting body (5.0) was observed in WB+ maize powder (MP) (1:1) supplement.

Yield: Highly significant variation was observed in yield of shaggy mushroom and it ranged from 125.0 to 170.0 g/packet (Table 1). The highest yield (170.0 g/ packet) was recorded in WB+RB (1:1) supplement, which was significantly higher to other treatments. The supplement WB+MP (1:1) performed poorly.

Effect of supplements on the size of fruiting body: Significant variation was observed in the size of fruiting body of shaggy mushroom grown on sawdust supplemented with different supplements. The length of stalk was highest in WB+RB (1:1), which was significantly higher to all other treatments. The shortest length of stalk was observed in WB. The diameter of stalk was highest in WB+RB (1:1), which was statistically similar to WB+MP (1:1) and the lowest diameter of stalk was observed in MP. The higher length of pileus was observed in WB+RB (1:1), which was a statistically similar to all other treatment except WB and MP. The highest diameter of pileus was observed in WB+MP,

which was statistically similar to WB+RB (1:1) and RB+MP (1:1) and the lowest diameter of pileus was observed in WB.

Table 1. Effect of different supplements on the number of fruiting body and yield of shaggy mushroom

Supplement	Number of fruiting body	Yield g/ packet
Wheat Bran (WB)	7.75 ab	153.0 b
Rice Bran (RB)	6.25bc	138.3 c
Maize Powder (MP)	6.25bc	125.0 d
WB+RB (1:1)	9.00a	170.0 a
WB+MP (1:1)	5.00c	139.5c
RB+MP (1:1)	6.00bc	146.5 bc
CV%	16.57	5.69

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Table 2. Effect of different supplements on the size of fruiting body of shaggy mushroom

Supplements	Length of stalk (cm)	Diameter of stalk (cm)	Length of pileus (cm)	Diameter of pileus (cm)
Wheat Bran (WB)	3.75c	1.37 c	3.62 b	1.52 b
Rice Bran (RB)	4.15 bc	1.22 cd	3.97 ab	1.40 b
Maize Powder (MP)	3.87 bc	1.10 d	3.52 b	1.62 b
WB+RB (1:1)	5.22 a	2.00 a	4.52 a	1.97 a
WB+MP (1:1)	4.37 bc	1.93 a	4.05 ab	2.12 a
RB+MP (1:1)	4.55 b	1.62 b	4.15 ab	1.90 a
CV%	10.48	8.83	10.29	8.79

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

Effect on nutritional composition of shaggy mushroom: The carbohydrate and protein content of shaggy mushroom was remarkably influenced by different supplements (Table 3). The highest percentage of carbohydrate (41.75%) was observed in the mushroom grown on MP supplemented sawdust, which was followed by RB+MP (1:1), WB+MP (1:1) and RB supplement. The lowest percentage of carbohydrate (36.13%) was observed in WB+RB supplement. The maximum protein content (29.75%) was observed in mushroom grown on WB + RB (1:1) supplemented sawdust followed by WB, RB+MP (1:1), WB+MP (1:1). The lowest protein content (27.25%) was observed in MP.

Fat, fiber and ash content of shaggy mushroom were not remarkably influenced by different supplement (Table 3). The lowest fat content (4.75%) was observed in MP and RB+MP (1:1) supplement and the higher fat content was recorded in WB, RB and WB+MP (1:1). The highest fiber content (21.75%) was observed in WB+RP (1:1), followed by WB+MP (1:1) and RB+MP (1:1). The lowest fiber content was recorded in

WB (19.00%). Comparatively higher ash content was observed in shaggy mushroom grown on WB+RB (1:1) and RB supplemented sawdust.

The difference in nutrient content of the same mushroom species might be due to the difference in nutrient content of substrates. The influence of substrates on nutrient content of other mushroom was reported by Sarker *et al.* (2007) and Qin (1989).

Table 3. Effect of different supplements on the nutrient content (%) of shaggy mushroom

Supplement	Carbohydrate	Protein	Fat	Fiber	Ash
Wheat Bran (WB)	37.00	29.50	5.00	19.00	7.00
Rice Bran (RB)	38.25	28.50	5.00	19.26	7.25
Maize Powder (MP)	41.75	27.25	4.75	19.75	6.50
WB+RB (1:1)	36.13	29.75	4.88	21.75	7.50
WB+MP (1:1)	39.50	29.25	5.00	21.50	7.00
RB+MP (1:1)	40.00	29.50	4.75	20.50	6.70
CV (%)	7.66	3.92	11.29	11.63	18.29

In a column, means followed by a common letter are not significantly different at 5% level by DMRT.

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Review Article

Oyster mushroom (*Pleurotus* spp.): A Friendly and Medicinal Mushroom

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Abstract

For centuries, people have enjoyed mushrooms for their flavor, texture and mystique. Eastern cultures have revered mushrooms as both food and medicine for thousands of years. Among the mushroom kingdom, Oysters are one of the most versatile mushrooms. They are easy to cultivate and common all over the world. The latin name *Pleurotus ostreatus* means "sideways oyster", referring to the oyster-like shape of the mushroom. They are found on hardwoods throughout the world in the spring and fall. The caps usually range between 5 to 25 cm (2 to 10 inches) and are shaped like a fan or an oyster. The caps are rolled into a convex shape when young and will flatten out and turn up as the mushroom ages. They are also very beautiful, coming in a broad spectrum of colors. They can be white, yellow, brown, tan, and even pink. They have a unique scent that is often described as sweet like anise or licorice (liquorice). Due to the wide range of activities this review summarizes various research findings conducted by different agencies worldwide about oyster mushroom.

Key words: *Pleurotus* spp., beta glucan, antioxidant, chitin, chitosan, pleuran.

INTRODUCTION

The Oyster mushroom (*Pleurotus ostreatus)*, first cultivated in Germany as a subsistence measure during World War I (Eger *et al.*, 1976) is now grown commercially around the world for food. In Japanese, Korean and Chinese cookery, oyster mushroom is frequently used as a delicacy. It is a new addition of food crop in Bangladesh, needs no or minimum cultivable agricultural land for growth. It grows well in useless land with shadow and inside the home. For millennia, mushrooms have been valued as edible and medical provisions for humankind. With the popularization of mushroom farming and/or industrialization, mushroom production worldwide continues to increase. Mushroom production can convert the huge lignocellulosic waste materials into a wide diversity of products (edible or medicinal food, feed and fertilizers), protecting and regenerating the environment. In addition, the mushroom production can generate equitable economic growth that has already had an impact at national and regional levels. The mushroom conversion has been named the 'non-green revolution' (Chang, 1999_a and Chang, 2005). However, the mushroom science is a relatively new applied science and the mushroom industry is still small compared to many plant crops, so the investment is limited. As a

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consequence, scientific research on mushrooms generally lags behind that of plant and animal (Sonnenberg, 2005).

Fig. 1. Different types of oyster mushroom.

In recent years, some edible mushrooms are used as health foods, as well as a source for pharmaceutical compounds. In fact, these functional mushrooms are a source of biologically active substances with therapeutic effects due to their immunomodulating, anticancer and antivirus properties (Wasser and Weis, 1999), among others. Many commercially available mushrooms exhibit free radical scavenging, reducing power, chelating effects on metal ions, and antioxidant properties (Mau *et al.*, 2002 and Yang *et al.*, 2002).

Considering these situations, mushrooms can play an important role in improving the nutritional status of the population. Mushrooms are recognized as an alternative source of good quality protein and are capable of producing the highest quantities of protein per unit area and time from the worthless agro wastes (Chadha and Sharma, 1995). Also they are the source of extraordinary vigor, power and vitality and are used in the preparation of many continental dishes. Dietary mushrooms provide wide variety of medicinal properties and they are active against certain life threatening diseases. It is now established that

mushrooms are good source of high quality proteins and minerals (Pathak *et al.*, 1998). The nutritional value of mushroom protein lies between meat and vegetables (Bhal., 1984). They are low calorie food with very little fat and sugars and without starch and cholesterol (Pathak *et al.*, 1998). Mushrooms are not only sources of nutrients but also have been reported as therapeutic foods. Mushroom of *Pleurotus* spp. are also rich in medicinal values and useful in preventing disease such as hypertension, hypercholesterolemia (Khatun *et al.*, 2007 and Choudhury *et al.*, 2008) hyperglycemia and different types of cancer (Nayana and Janardhanan, 2000). In China, oyster mushroom is indicated for joint and muscle relaxation (Yang and Jong., 1989).

Cultivation of oyster mushroom

Though often grouped with vegetables, mushrooms are fungi. So, all of them grow from microscopic spores, not seeds. Plants that grow from spores are called fungi. Mushrooms belong to two subdivisions of fungi, Basidiomycetes and Ascomycetes, with most belonging to the Basidiomycetes subdivision. A mushroom is often defined as the fruiting body of a macrofungus. The fruiting body refers to the spore-producing organ of a fungus, which is formed by the mycelium and consists of a stem or stalk crowned by a cap called pileus ($Anon_a$).

Spores, dropped from mature mushrooms and collected in the nearly sterile environment of a laboratory, are used to inoculate grains or seeds to produce a product called spawn, which is equivalent of seed. Mushrooms have no chlorophyll and must get all their nutrients from organic matter in the growing medium. The medium, called compost is scientifically formulated from various materials such as straw, corncobs, cottonseed and cocoa seed hulls, gypsum and nitrogen supplements. The compost is pasteurized and placed in large trays or beds. The spawn is worked into the compost and the growing takes place in specially constructed houses, where the grower can regulate the crucial aspects of heat and humidity ($Anon_a$).

In several weeks, the compost becomes filled with the root structure of the mushroom, the network of lacy white filaments called mycelium. A casing layer of wet peat moss is spread over the compost. Eventually, tiny white protrusions form on the mycelium and push up through the peat moss, termed pinning by the growers. The pins continue to grow into the mushroom caps, which are actually the fruit of the plant. It takes 17 to 25 days to produce mature mushrooms after the peat moss is applied. The entire process from the time the grower starts preparing the compost until the mushrooms are harvested and shipped to market takes about four months ($Anon_a$). Harvested mushrooms are set in carts, refrigerated and then packaged and shipped quickly to supermarkets, food processors and restaurants for consumers to enjoy in a wide range of recipes, prepared foods and signature dishes of world-class chefs (Anon, 2000).

Cultivation of oyster mushroom is a very simple procedure: Cultivation of the Oyster mushroom, *Pleurotus* spp., has increased greatly throughout the world during the last few decades (Chang, 1999_b and Royse, 2002). In 1997, it accounted for 14.2 % of the total world edible mushroom production. Its popularity has been increasing due to its ease of

cultivation, high yield potential and high nutritional value (Banik and Nandi, 2004). Although commonly grown on pasteurized wheat or rice straw, it can be cultivated on a wide variety of lignocellulosic substrates, enabling it to play an important role in managing organic wastes whose disposal is problematic.

Oyster mushroom cultivation is a very simple procedure in the case of log cultivation because it does not involve sophisticated equipment. However, despite its simplicity, large-scale cultivation on natural logs is not often used due to long incubation periods, low yields and environment-dependent production if conducted outdoors. Yields of *P. ostreatus* fruiting bodies vary with the species of trees used and range from 21 % biological efficiency (BE) for beech wood to 3 % BE for alder wood (Pavlik, 2005).

Broadleaf, hardwood, sawdust and straw-based substrates with added supplements are more often used in commercial production. In this case, these artificial substrates must be pretreated, mainly for elimination of contaminants, and handled in a clean environment. There are different methods of cultivation like shelf, bag, bottle, tray, jar, grid-frame, wall-frame and others (Stamets, 2000). In practice, the most used are bag, bottle and shelf cultivation (Anon, 2003).

Pleurotus spp. can also colonize and produce mushrooms on pretreated conifer (*Pinus* spp.) wood chips but they do not always readily colonize non-pretreated conifer wood, due to the presence of inhibitory components (Croan, 2004). Some strains can, however, be adapted for cultivation on conifer-sawdust-based substrates (Ruan *et al.*, 2006). *Pleurotus* spp. can also be cultivated on wood waste or unused wood residues associated with harvesting or thinning operations, which can enhance economic returns needed to support ecosystem management (Croan, 2000).

Cultivation of oyster mushroom is influenced by various factors: Cho *et al.* (2003) discovered that inoculation of pure *P. ostreatus* mycelium cultures with strains of fluorescent *Pseudomonas* spp., isolated from the mycelial plane of commercially produced mushrooms, promoted the formation of primordia and enhanced the development of the basidiomata. These results strongly suggest that inoculation of the mycelium with specific bacteria may have beneficial applications for mushroom production.

Qu *et al.* (2006) demonstrated the influence of heavy metals in substrates on *P. eryngii* primordial formation, fruiting body development and BE. Heavy metal (As, Hg, and Cd) supplementation decreased average growth yields and BE of *P. eryngii*, whereas Pb supplementation improved both parameters. Irradiation by red and green light stimulated vegetative growth of *P. ostreatus* mycelium and shortened the substrate colonization and fructification time. The increased fruiting body yield in irradiated cultures reached 36–51 % (Poyedinok *et al.*, 2003). The cytolytic protein ostreolysin, isolated from *P. ostreatus* fruiting bodies, was specifically expressed during fruiting initiation, suggesting its involvement in fruiting body formation. Nasim *et al.* (2001) found that malt extract agar (MEA) provided faster *P. ostreatus* mycelial growth rates than did Murashige and

Skoog's (MS) medium and potato dextrose agar (PDA). The slowest growth was observed on PDA medium. Addition of fructose to basal medium, supported higher mycelial yields than the addition of glucose. Yeast extract as the nitrogen source proved better than peptone when monosaccharides were used as the sole carbon source (Wu *et al.*, 2003).

Mycelium production on lignocellulosic substrates has also been investigated. Amongst seven mushroom cultivation substrates, the mycelial extension rates were highest on cotton gin-trash, peanut shells and poplar sawdust. Supplemented oak sawdust and olive mill waste were poor substrates for most species examined, while almost all strains performed adequately on corn cobs (Zervakis *et al.*, 2001 and Gregori *et al.*, 2006). Banana leaf waste was a better substrate than banana pseudostem waste in the production of extracellular enzymes by *P. ostreatus* and *P. sajor-caju* and is a potential alternative to other agrowaste substrates. This is in agreement with Zhang *et al.* (2002) who reported that *P. sajor-caju* grew faster and provided better yields on ground straw than on chopped straw.

Chemical composition of mushroom depends on type of substrate: *P. ostreatus* and *P. sajor-caju* exhibited higher ash content when cultivated on rice straw than when cultivated on banana straw, and *P. sajor-caju* also showed higher moisture and fiber content when cultivated on rice straw (Bonatti *et al.*, 2004). When cultivating *P. ostreatus* on corn and pumpkin straw, the substrate had no effect on the nitrogen content and amino acid profile of the fruiting bodies; however, the nitrogen content increased from the first harvest to the third harvest (Mendez *et al.*, 2005).

Mandeel *et al.* (2005) cultivated *Pleurotus* spp. on various lignocellulosic wastes supplemented with fresh chicken manure. The highest biological efficiency (BE) was noted on cardboard with both *P. columbinus* (134 %) and *P. ostreatus* (117 %). Experiments conducted by Baysal *et al.* (2003), which involved cultivation of *P. ostreatus* on waste paper with addition of chicken manure, peat and rice husks, showed that increasing the amount of rice husks added to the substrate accelerated spawn running, pinhead formation and fruiting body formation. The chemical analysis of fruiting bodies indicated that *P. ostreatus* cultivated on spent grain substrate had a higher nutritional value than those grown on other types of substrates (Wang *et al.*, 2001).

Quality of mushroom depends on cultivation method: The nature of the substrate as well as the cultivation method affects the expression of lignocellulolytic enzymes. The study conducted by Elisashvili *et al.* (2008) revealed that solid-state fermentations (SSF) of tree leaves by *Pleurotus* spp. was favourable for laccase and manganese peroxidase (MnP) production. Furthermore, coculturing can be an effective method for biopulping and improvement of lignin degradation (Watanabe *et al.*, 2003 and Chi *et al.*, 2007). Chi *et al.* (2007) demonstrated that coculturing *P. ostreatus* with *Ceriporiopsis subvermispora* significantly stimulated lignin degradation when compared to monocultures. Laccase production and MnP activity were stimulated in cocultures of *P. ostreatus* with *C. subvermispora* or *Physisporinus rivulosus* and a change in the isoform composition of

those enzymes was also observed. These studies show that the cultivation method can have drastic effects on the production of valuable substances by *Pleurotus* spp.

Recent studies on mushroom polysaccharides have demonstrated many interesting biological activities, which are described later in this review. The production of *Pleurotus* spp. mycelial biomass and valuable polysaccharides in submerged liquid fermentation (SLF) depends on the species used, growth parameters, growth timing and their nutritional requirements (Kim *et al.*, 2002 and Rosado *et al.*, 2003).

Production of mushroom: China is the world's largest producer of mushrooms with 32% of output. The United States follows China with 16% World output. The U.S. commercial mushroom industry began in the early 1900s in Kennett Square, PA. Pennsylvania (53% of the 1999-2001 U.S. output), California (15%) and Florida (5%) are the top producing states, although 30 other states also report production.

P. ostreatus as an edible mushroom

Oyster mushrooms are also a popular edible. They have a nutty, subtle flavor that goes well in soups, stews, and sauces. Fortunately *P. ostreatus* is one of the easiest species to cook. Although they go well in many dishes, the most common way to cook oyster is a simple saute or stir fry. Brown them in oil with herbs and spices of choice. Mushrooms are a flavorful and nutritious food group. *P. ostreatus* is extremely delicious as well as conferring various health-giving properties and benefits. They are good sources of B-Vitamins: Thiamine, Riboflavin and Niacin. They contain all the essential amino acids. Mushrooms have also been used for thousands of years as some of the most effective, yet benign, of many plants that formed the oriental herbal tradition. The dried Oyster mushrooms are said to be high in iron, so they are potentially good blood builders.

Nutritional components

Mushrooms have been part of our human diet since time immemorial. They were used as food even before man understood the use of other organisms. Undoubtedly, mushrooms were one of man's earliest foods, and they were often considered an exotic and luxurious food reserved for the rich. Today mushrooms are food for both the rich and the poor. Now it is believable that mushroom eaters have a better nutrient profile than do those who do not eat mushrooms (Feeney, 2003).

Mushrooms are low in calories and fat, but have higher protein, than most vegetables and are good sources of vitamins. Nutrient composition and biologically active compounds can vary widely due to differences in mushroom type/variety, the substrate or growing medium, the developmental stage and age of the mushroom. The nutrient content of fresh mushrooms also is related to moisture content, which depends on conditions during cultivation. Mushrooms are approximately 92% water by weight, with protein, fat, carbohydrate, fiber and ash making up the remaining 8% of the dry weight (Matilla *et al.*, 2002) Carbohydrates represent approximately half of the weight of the dry matter. Carbohydrates in mushrooms include polysaccharides such as glucans, mono-and

disaccharides, sugar alcohols, glycogen and chitin, the major cell wall component. Mushrooms contain a variety of amino acids, including the essential amino acids. Although white mushrooms contain less than half a gram of total fat, the three major types of fatty acids are present including the essential fatty acid, linoleic acid.

Potassium, phosphorus, and magnesium account for most of the mineral content. Mushrooms also contain some calcium, iron, copper, zinc and selenium. However, only copper, selenium and potassium are present in nutritionally significant amounts. Most fungi produce ergosterol, a precursor to vitamin D_2, under sunlight or ultraviolet irradiation. They are a good source (10% Daily Value) or excellent source (20% Daily Value) of selected vitamins and minerals, riboflavin, niacin, pantothenic acid, potassium, copper, and selenium ($Annon_b$). Some nutritional components of *P. ostreatus* analyzed by Regula, and Siwulski, (2007) plotted below (Table-1).

Table 1. The energy value, kcal/100 g, and contents of macrocomponents, g/100 g dry matter, of dried *P. ostreatus*. Minerals and toxic metals of dried *P. ostreatus* mg/kg dry matter

Energy value and name of macrocomponents	Amount in dried *P. ostreatus* gm/100 gm	Name of microcomponents	Amount in dried *P. ostreatus* mg/Kg
Energy value	345.±1.84a	Iron	68.6 ±5.50
Water	10.6 ±0.28a	Coper	12.9 ±1.36
Protein	15.7 ±0.37	Zinc	109.6 ±0.89
Fat	2.66 ±0.06	Magnesium	1 289 ±20.4
Carbohydrates	64.1 ±0.01a	Calcium	27.6 ±0.15
Ash	7.04 ±0.15	Potassium	33 120.±191
Soluble dietary fiber	2.01 ±0.33a	Sodium	133.7 ±21.4a
Insoluble dietary fiber	39.8 ±0.55	Lead	0.000 ±0.000
		Cadmium	0.70 ±0.05
		Mercury	0.08 ±0.003

Evaluation of fibers of mushroom: The fiber of plants is cellulose, lignin and hemicellulose (Albershiem, 1976). The fiber of mushrooms is chitin (Robson, 1999). Often times chitin is referred to as "cellulose-like." Chitin and cellulose do have many chemical and mechanical properties in common, but they are also quite different from each other (Noller, 1951). Chitin is also the material that makes up the horny shells of crabs, lobsters, shrimp, and other arthropods.

Chemically, cellulose is poly-ß-(1-4)-D-glucose, while chitin in poly-ß-(1-4)-2-acetomindo-2-deoxy-D-glucose or more simply poly-N-acetyl-D-glucosamine (Windholtz, 1983). (**Fig. 2**). If chitin loses its acetyl groups through hydrolysis, it is called chitosan (Geddes, 1949).

Dietary fibers are not digested and so has no proper nutritional value, however, the fact that they go through the gut without being digested means that they can carry things through that are not good. Experiments with oats also show that they move other unhealthy materials out of the human system (Katz, 2001). While chitin and other fibers

have no nutritional value, they have great dietary, or health value. Like cellulose, chitin can not be digested in our gastrointestinal track, but also like cellulose, chemical digestion is possible. However, unlike glucose, the product of the chemical digestion of cellulose, glucosamine, the product of chemical digestion of chitin, has more nutritional value than just calories.

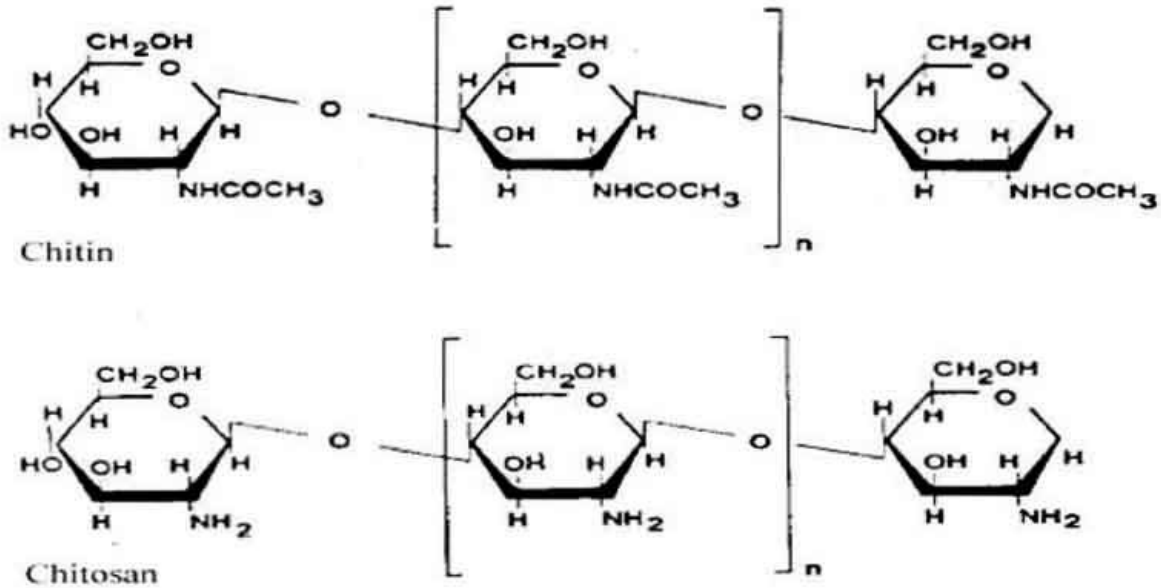

Fig. 2. Chemical structure of chitin (top) and chitosan (bottom).

Both chitin and chitosans from shellfish have been studied as dietary fiber. Fibers help to clean digestive tracks and reduces the chances of cancer and also heart disease and stroke. The studies suggest that chitin and chitosans are excellent hypocholesterolemics that is they reduce cholesterol. It is by the reduction in cholesterol that they reduces heart disease and strokes. It has been suggested that chitosans and to a lesser degree chitin may chelate some undesirable nutrients. Experiments suggest that chitosans capture cholesterol and bile in the intestines and carry them out so that the body can not absorb or reabsorb them. Dietary fiber supplements are used, especially to elderly and people on restricted diets who may have problems with constipation. Mushroom fiber reduces constipation and they have also the capability for reducing weight.

A new use of mushroom citosan that is not food is, mushroom wastes have been converted to chitosan and used as dressing on human wounds (Su *et al.*, 2004), and it appears to be very effective.

ß -glucan: Oyster mushroom is a source of a natural immunomodulating agent ß -glucan (**Fig. 3**), which is called "*pleuran*". In order to act actively in the human organism, this molecule must be isolated from the oyster mushroom and purified. ß -glucan is contained in the cell walls of mushrooms, where it is firmly bound to other molecules responsible for the strength and shape stability of the cells. Besides ß -glucan, the cell wall also contains other saccharides and proteins that are linked and bound together to form a solid structure. ß -glucan falls into a group of immunomodulating substances called PAMP

(*pathogen associated molecular patterns*). These molecules are non-specifically identified by the components of the immune system as the structures against which the immune response must be activated.

The effect of ß -glucan is realized through a direct contact with immunocompetent cells, macrophages. These cells contain several receptors (toll-like receptor, Dectin-1) on their surface to which the ß -glucan molecule is selectively bound, creating a bond that may be compared to a "key and lock" system. Not every key will open every lock – similarly, a specific pure molecule that "fits in" must be bound to the receptor. Only then, a cascade of signals is activated, the result being active immune response of the whole organism. Biological availability and immunomodulating effect of ß -glucan are directly dependent both on the structure of particular beta-glucan and the purity of obtained molecule. So it is unwise to use oyster mushroom powder as a mean to support immunity, because scientific evidence proves it cannot possess such properties.

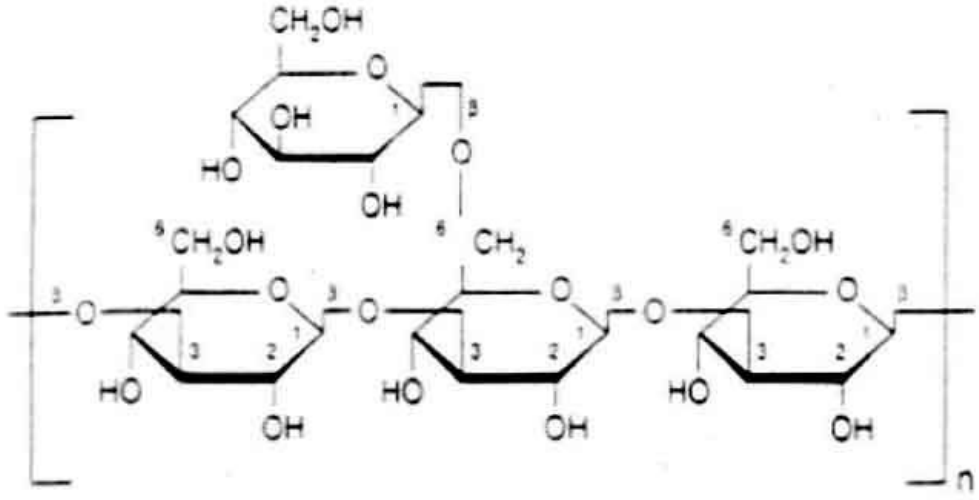

Fig. 3. ß -(1,3/1,6)-D-glucan.

Potassium: It's role as an essential mineral is well established. Potassium helps maintain normal heart rhythm, fluid balance, muscle, and nerve function. The Dietary Approaches to Stop Hypertension (DASH) Trial and the subsequent DASH-Sodium Trial looked at the effect of eating patterns rich in fruits, vegetables, fiber and low fat dairy products on the reduction of morbidity and mortality related to high blood pressure in the general population (Obarzanek *et al.*, 2001).

Selenium: Selenium, a trace element, is a constituent of selenium-containing proteins. Selenium is incorporated into proteins as the amino acids selenomethionine and selenocysteine. Selenomethionine cannot be synthesized by humans and is initially synthesized in plants. It is randomly incorporated in place of methionine in a variety of proteins obtained from plant and animal sources (Anon., 2000). Scientists have known for some time that selenium is important for proper growth and reproduction in animals. In humans, selenium functions largely through association with selenoproteins, several of

which are oxidant defense enzymes, to protect tissues against oxidative stress, regulate thyroid hormone metabolism, and help regenerate ascorbic acid from its oxidized metabolites (Greger, 2001).

Mushrooms provide more selenium than other fruits and vegetables in the produce category. Some studies suggest the possibility that intakes of selenium above those needed to maximize selenoproteins may have an anticancer effect in humans.

Table 2. Comparison of Nutrient contents of dried *P. ostreatus* (g/100 g)

Elements	Bangladesh	Poland
Carbohydrate	037.8 ± 2.5	64.1 ± 0.01
Protein	23.91 ± 2.0	15.7 ± 0.37
Lipid	04.6 ± 0.26	2.66 ± 0.06
Dietary fiber	24.34 ± 1.8	Total = 41.8 Soluble = 2.01 ± 0.33 Insoluble = 39.8 ± 0.55
Ash	9.36 ± 0.5	7.07 ± 0.15

Analysis in Bangladesh done by Alam *et al.* (2008) and in Poland by Regula and Siwulski (2007)

Table3: Comparison of some Mineral contents of dried *P. ostreatus* (mg/100 g)

Minerals	Bangladesh	Poland
Iron (Fe)	55.45 ± 5.2	68.6 ± 5.50
Zinc (Zn)	26.56 ± 2.2	109.6 ± 0.89
Magnesium (Mg)	16.39 ± 2.1	1289 ± 20.4
Calcium (Ca)	35.9 ± 3.8	27.6 ± 0.15

Analysis in Bangladesh done by Alam *et al.* (2008) and in Poland by Regula and Siwulski (2007)

Medicinal properties

Studies conducted over the past 30 years, mostly in Asia, have provided data suggesting that mushrooms or substances extracted from mushrooms may aid in the treatment of certain types of cancer, boost the immune system and reduce the risk of coronary heart disease. Currently, U.S. researchers are studying the potential role of nutrients such as selenium and other compounds in mushrooms in the treatment and prevention of chronic diseases including breast and prostate cancer (Mattila *et al.*, 2001).

Many cultures have used mushrooms as both food and medicine, but the use of mushrooms as a functional or medicinal food is most notable in the East. The use of mushrooms to maintain health was recorded formally as early as 100 AD in China. Recent studies on various *Pleurotus* spp. have shown a number of therapeutic activities, such as antitumour, immunomodulatory, antigenotoxic, antioxidant, anti-inflammatory, hypocholesterolaemic, antihypertensive, antiplatelet-aggregating, antihyperglycaemic, antimicrobial and antiviral activities.

Mushrooms contain a wide variety of bioactive molecules including terpenoids, steroids, phenols, nucleotides and their derivates, glycoproteins, and polysaccharides (Borchers *et*

al., 1999 and Mizuno *et al.*, 1995). Much of the work on the anti-tumor activity of mushrooms has concerned the polysaccharides, which appear to be potent anti-tumor active compounds. Although many mushrooms contain several polysaccharides that inhibit tumor growth, most are the branched (1-3)-β-D-glucans. Polysaccharides in whole mushrooms and isolated mushroom compounds modulate immune system and potentially exert antitumor effects (Borchers *et al.*, 1999).

Antitumour effect: Polysaccharides, proteins and other substances of *Pleurotus* spp. possess antineoplastic activities *in vitro* and *in vivo*. Various crude extracts of *Pleurotus* spp. have been shown to possess relatively strong antitumour activities. Methanol extracts of *P. florida* and *P. pulmonarius* fruiting bodies significantly reduced solid tumours in mice (Jose and Janardhanan, 2000 and Jose *et al.*, 2002). *P. ostreatus* mycelium extract, alone and combined with the chemotherapeutic agent cyclophosphamide, inhibited *in vivo* tumour growth in mice. The combined administration of the extract with cyclophosphamide decreased the degree of leukopenia compared to administration of cyclophosphamide alone (Meerovich *et al.*, 2005). A water extract of *P. ostreatus* exhibited the most significant cytotoxicity by inducing apoptosis of human carcinoma cells, when compared to many other mushroom extracts. It has been suggested that the active compounds in the extract were water-soluble proteins or polypeptides (Gu and Sivam, 2006). Antitumour properties have also been demonstrated for *Pleurotus* spp. proteins, proteoglycans, and DNA. A lectin isolated from *P. ostreatus* potently inhibited growth of sarcoma and hepatoma in mice and prolonged their lifespan (Wang *et al.*, 2000).

Furthermore, two ribonucleases isolated from *P. sajor-caju* and *P. ostreatus* fruiting bodies exhibited antiproliferative effects on tumour and leukaemia cell lines (Ngai, and Ng., 2004. and Xia *et al.*, 2005). Another protein, eryngeolysin, isolated from *P. eryngii* fruiting bodies, exhibited cytotoxicity against leukaemia cells (Ngai and Ng, 2006). Water-soluble proteoglycans were purified from *P. ostreatus* mycelium and exerted antitumour activity in sarcoma-bearing mice. Proteoglycans injected into mice reduced the number of tumour cells by cell cycle arrest (Sarangi *et al.*, 2006). Moreover, DNA isolated from *P. ostreatus* fruiting bodies administered to mice with solid Ehrlich carcinoma significantly increased the lifespan of mice (Shlyakhovenko *et al.*, 2006).

Immunomodulatory effect: several compounds from *Pleurotus* spp. with immunostimulatory activities on humoral and cell-mediated immunity have been isolated. Glucans isolated from *P. florida* fruiting bodies activated the phagocytic response of mouse macrophages *in vitro* (Rout *et al.*, 2005) and significantly induced the proliferative response as well as phagocytic activity of fish leukocytes *in vitro* (Kamilya *et al.*, 2006). Proteoglycans from *P. ostreatus* mycelia exerted immunomodulatory effects by elevating mouse natural killer cell cytotoxicity and by macrophage stimulation (Sarangi *et al.*, 2006). DNA isolated from *P. ostreatus* fruiting bodies stimulated mouse natural killer cytotoxic activity *in vitro* (Shlyakhovenko *et al.*, 2006).

Antimitogenic effect: Antimitogenic effects of *Pleurotus* spp.-derived compounds on immune cells have also been reported. A ribonuclease isolated from *P. sajor-caju* fruiting

bodies exerted antiproliferative effect on murine splenocytes (Ngai and Ng, 2004.), while eryngeolysin from *P. eryngii* inhibited the stimulated mitogenic response of murine splenocytes (Ngai and Ng, 2006).

Antioxidant effect: Antioxidant compounds prevent oxidative damage related to aging and diseases, such as atherosclerosis, diabetes, cancer and cirrhosis. Mushrooms that contain antioxidants or increase antioxidant enzyme activity may be used to reduce oxidative damage in humans (Yang *et al.*, 2004). An extract from *P. cornucopiae* possessed the most effective antigenotoxic and bio-antimutagenic activities when tested on *Salmonella typhimurium* and *Escherichia coli* (Filipic *et al.*, 2002). *P. cornucopiae* extracts significantly reduced H_2O_2-induced DNA damage in Chinese hamster lung cells (El Bohi *et al.*, 2005). Methanol extracts of *P. ostreatus* and *P. cystidiosus* fruiting bodies possessed antioxidant, reducing power, radical scavenging and iron chelating activities (Yang *et al.*, 2004). Methanol extracts of *P. florida* and *P. pulmonarius* fruiting bodies showed similar antioxidant activities (Jose and Janardhanan, 2000 and Jose *et al.*, 2002). *P. citrinopileatus* fruiting body extracts have shown antioxidant activities *in vitro* and in hyperlipidaemic hamster rats (Hu *et al.*, 2006). Pleuran, a β-glucan isolated from *P. ostreatus*, had a positive effect on the antioxidant status of rats and decreased precancerous lesions induced in rat colon (Bobek and Galbavy, 2001).

Anti inflammatory effects: Studiy shows that methanol extracts of *P. pulmonarius* and *P. florida* fruiting bodies decreased induced paw oedema in mice and ameliorated acute and chronic inflammation (Jose *et al.*, 2002 and Jose *et al.*, 2004). Pleuran has also been shown to possess anti-inflammatory effects by exerting antioxidant and immunomodulatory effects on rats with induced colitis (Nosálová *et al.*, 2001). Hypersensitive immune responses, such as inflammation in delayed allergy, were suppressed by an ethanol extract of *P. eryngii*. It exhibited anti-allergic activity after oral or percutaneous administration to mice with oxazolone-induced type IV allergy (Sano *et al.*, 2002).

Antihypercholesterolemic effects: Lowering of the serum cholesterol and lipid levels plays a significant role in the prevention of arthrosclerosis and other nutrition-related diseases. This has been attributed to their high dietary fiber levels (Bobek *et al.*, 1998) and other components such as eritadenine, guanylic acid and ergosterol (Anderson *et al.*, 1988). Oyster mushroom has lots of bioactive compounds with hypocholesterolaemic activities, which includes polysaccharides, mevinolin and other statins (Gunde-Cimerman., 2001). Statin drugs reduce bad cholesterol (LDL-c) by stimulating receptors in the liver to clear the cholesterol from the body. Studies have shown a link between consuming *P. ostreatus* and a lowering of cholesterol levels, no doubt due to the stains they produce. It has recently been reported that *P. citrinopileatus* fruiting body extracts exerted antihyperlipidaemic effects. Serum triglyceride and total cholesterol levels were lowered in hyperlipidaemic rats supplemented with the extracts, while high-density lipoprotein levels were significantly increased (Hu *et al.*, 2006_a). Similar effects were noted when powdered *P. ostreatus* fruiting bodies were fed to hypercholesterolaemic rats (Hossain *et al.*, 2003).

Antihypertensive effects: *Pleurotus* spp. possess blood pressure lowering effects. Recently in a study, *P. cornucopiae* has exhibited antihypertensive activity; this might be due in part to D-mannitol, which inhibits angiotensin I converting enzyme (Hagiwara *et al.*, 2005). A methanol extract of *P. florida* fruiting bodies significantly inhibited platelet aggregation. The antiplatelet-aggregating activity, along with the anti-inflammatory activities, suggests its potential therapeutic use against hypertension.

Hepatoprotective effects: Study showed some hepatoprotective activity of oyster mushroom (Mishra and Singh., 2010 and Sumy *et al.*, 2010). It is evident that addition of oyster mushroom to the diet effectively reduced cholesterol accumulation in serum and liver of adult human (Opletal *et al.*, 1997 and Jayakumar *et al.*, 2006). It increases the levels of reduced glutathione in the liver and stimulates the activities of catalase and glutathione peroxidase in the liver (Pathak *et al.*, 1998). In a study it was observed that supplementation of 50 grams of *P. ostreatus* as Ifter item significantly reduced plasma ALT and AST at Ramadan (Choudhury *et al.*, 2009) but the reduction rate of ALT was more marked in comparison to AST. On the other hand it was observed that the reduction rate of both ALT and AST was more prominent in case of male than that of female (Choudhury *et al.*, 2010).

Antihyperglycemic effects: Antihyperglycaemic effects were demonstrated with a water-soluble polysaccharide from *P. citrinopileatus* fermentation broth. The polysaccharide was effective in lowering blood glucose levels in diabetic rats (Hu *et al.*, 2006_b).

Antimicrobial effects: Various studies suggest antimicrobial and antifungal activities of *Pleurotus* spp. extracts and isolated compounds. Crude extracts of *P. ostreatus* from fermentation broth inhibited Gram-positive, Gram-negative bacteria and *Aspergillus niger* fungi (Gerasimenya *et al.*, 2002). Hexane-dichloromethane extract containing *p*-anisaldehyde of same species was effective against *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Aspergillus niger* and *Fusarium oxysporum* (Okamoto *et al.*, 2002). Eryngin, an antifungal peptide and Eryngeolysin, a haemolysin of *P. eryngii* exibated activity against *Fusarium oxysporum*, *Mycosphaerella arachidicola* and *Bacillus* spp. (Wang, and Ng., 2004., Ngai, and Ng., 2006). 12 kDa ribonuclease of *P. sajor-caju was active against Fusarium oxysporum*, *Mycosphaerella arachidicola*, *Pseudomonas aeruginosa* and *Staphylococcus aureus* (Ngai and Ng, 2004.).

Antiviral effects: Various active agents from *Pleurotus* spp. extracts direct or indirect antiviral effects as a result of immunostimulatory activity (Brandt, and Piraino., 2000). Inhibitory activity against human immunodeficiency virus (HIV)-1 reverse transcriptase has recently been demonstrated for *P. sajor-caju* and *P. pulmonarius* hot water extracts (Wang *et al.*, 2007). Anti-HIV activity was also demonstrated for a ubiquitin-like protein isolated from *P. ostreatus* fruiting bodies (Wang, and Ng, 2000). Zhang *et al.* (2003) and Zhang *et al.* (2004) demonstrated that *P. tuber-regium* sclerotia, exert antiviral activities against herpes simplex virus type 1 and type 2. The effect is presumably elicited by the

binding of sulphated ß-glucans to viral particles, thus preventing them from infecting the host cells.

Adverse effects of taking mushroom

Food supplements might further be described as natural things that are used to prevent or at least lessen the chance of pain and disease. Unlike true medicines, they derive their benefit from promoting natural body functions and not from selective poisoning. That is not to say that they can never be poisonous. For example, high doses of niacin and other vitamins are poisonous (Windholtz, 1983), but food supplements do not derive benefit from the properties that make them poisonous.

Ostreolysin is a 15 kDa cytolytic protein found in considerable amounts in oyster mushrooms (*P. ostreatus*). Ingestion of large quantities of these edible mushrooms can result in adverse reactions, and at least some of these effects can be attributed to ostreolysin. This protein is able to permeabilize erythrocytes and other cells at sub-micromolar concentrations, acting by a colloid-osmotic mechanism and inducing the formation of a membrane pore with a hydrodynamic radius of 2 nm. Its binding to and subsequent permeabilization of the membrane depend on interaction with cholesterolenriched raft-like membrane domains. When injected intravenously into rodents, ostreolysin induces cardiorespiratory arrest and is lethal for mice with an LD50 of 1170 µg/kg. In rats, it induces a transient increase in arterial blood pressure, followed by a progressive drop of blood pressure, associated with noticeable bradicardia and myocardial ischemia. Continued drop of blood pressure is accompanied with ventricular extrasystoles, related to marked hyperkalemia. Moreover, sub-micromolar concentrations of ostreolysin induce a concentration-dependent increase in aortic ring tension (Sep and Framge, 2009).

Oyster mushrooms contain small amounts of arabitol, a sugar alcohol, which may cause gastrointestinal upset in some people. Some people are allergic to their spores while others may experience an upset stomach. In this situation it is wise to try a small amount first to see how body reacts.

Removal of pollutant from environment

Oyster mushrooms are one of the prime candidates for breaking down petroleum-based and hydrocarbon-based contaminants and pesticides. A project using fungal technologies took place after a diesel fuel spill near Bellingham, Washington, observed that oyster mushroom removed oil spill from soil. Subsequent laboratory tests found virtually no toxic oil residue in either the soil or the mushrooms, the result of enzymes and acids that the fungi release that break down such molecular complexes. This finding is especially significant because hydrocarbons are the basis for many other toxic industrial products, including most pesticides and herbicides. But the exciting part of the story was that, after the mushrooms become mature, flies came in and laid eggs in them. Maggots appeared, birds flew in, and other small mammals began to eat the mushrooms and the maggots. The birds and animals carried in seeds, and plants started growing. The mushrooms

initiated a process that led to rapid habitat recovery. The polluted pile of dirt was transformed into an ecosphere of life (Ausubel K., 2004).

In the Gulf of Mexico there occurred a worst disaster by leaking huge amounts of oil and toxic chemical into and sprayed on the Gulf, making the environment toxic to all life in September 2010 (**Fig. 4**). But it was possible to save individual animals by bioremediation projects using oyster mushrooms.

Fig. 4. Bioremediation is the only way to address the toxicity of this mix of Corexit and the oil (Photo courtesy of Williams, 2010).

Fig 5. Oyster mushrooms producing on oil contaminated soil (10,000-20,000 ppm) (Photo courtesy of Susan Thomas).

Fig. 6. Oyster mushroom fruiting from burlap over toxic spawn inoculated soil (Photo courtesy of Robert Rawson).

There were three possible methods (1) making burlap sacks filled with oyster mushroom spawn, spent oyster mushroom compost and substrate, (2) making cardboard tubes with mushroom spawn and staking them into oily ground and (3) broadcasting spawn in rows of wood chips, hay, and dead vegetation.

All three methods utilized oyster mushrooms because they grow easily, were good for bioremediation, and were fairly easy to obtain. When mushrooms grow they make an intricate network of thread-like cells called mycelium. These mycelium secrete powerful enzymes that can break down toxins into less toxic and non toxic compounds. After the mycelium finish fruiting (producing mushrooms) microbial bioremediation can be applied to complete the breakdown of the simple toxins (**Fig. 5 & 6**). If conditions for growing are good, bioremediation can occur in days to weeks. Under ideal conditions, mushrooms (fungi) are capable of generating enormous mycelia mats spreading over thousands of acres of land. (Williams, 2010).

Poisonous mushroom

Poisonous mushrooms are often referred to as "toadstools," but this is a folk name that has no precise meaning. Although Oyster mushroom has a number of look-alikes, (including *Crepidotus and* Lentinus spp.), but none are dangerous. They may, however, be woody or unpleasant- tasting. There are lots of other mushrooms which may be deadly poisonous. There are three most dangerous groups of fungi. These are amanitas, the false morels and a catch-all category known as little brown mushrooms (**Fig. 7**). Mushrooms in these groups cause virtually all the fatal mushroom poisonings. With amanitas alone accounting for 90 percent of mushroom-related deaths.

Oyster Amanitas False morels Little brown mushroom

Fig. 7. Differentiation of oyster mushroom from poisonous mushrooms.

DISCUSSION

Mushrooms are basically fungi, oyster mushroom is one of them which have a fleshy and spore-bearing fruiting body. They have been in use not only for consumption purposes, but also for medicinal purposes, since ages. There are over 14,000 types of mushrooms in the world, out of which about 3,000 are edible, about 700 have known medicinal properties and around 1400 have been recognized as poisonous. Today, mushrooms are eaten by people, for their flavor, texture as well as for the health benefits they accord. Mushrooms do certainly have enormous potential for feeding third world peoples. In the west, mushrooms are regarded as a luxury food. But in many developing countries of the world, mushroom can mean cash for the poor and a new source of nutrition.

Oyster mushrooms contain many things that fit the definition of food supplements. One kind of oyster mushroom may be richer in one of these materials while another kind will be richer in another. However, they are generally similar to each other in special food values. Oyster mushrooms are very good nutritionally and to explain what makes them so good. Any food with high nutritional value must be considered a health food.

Oyster mushrooms contain a number of enzymes which may participate in several clinical conditions such as tumor and cancer invasion and cardiovascular disorders. It has been known that enzyme therapy plays an important role in several clinical conditions such as in cancer treatment, malignant lymphoma and cardiovascular disorders as well as in the treatment of disorders of the peripheral and central nervous system, blood cells, metabolism of vitamin D and calcium, and reproductive system. These mushroom enzymes are thought to prevent oxidative stress as well as to inhibit cell growth in several diseases. Furthermore scientists are looking at ways of combating extreme tiredness designated as chronic fatigue syndrome caused by viral infections, hepatitis C and HIV. It could open the new modalities of treatment option for various diseases due to the presence of a lot of the biological active components in mushrooms.

Oyster mushroom has proven positive effects on the human body and spirit. Its main effects are based on active substances - beta glucans, which have the ability to activate cells for natural immunity to the organism. In addition, oyster mushroom contains a variety of vitamins B, D, C, K, proteins, sterols, fatty acids and some trace elements of

chromium, copper, iron, iodine, sodium, selenium and zinc. Oyster is also an important source of natural products from the statins (lovastatin, mevinolin), which protects against hardening of the blood vessel wall and has a positive effect on hypercholesterolaemia. Oyster mushroom is an important source of fiber, which consists of basic polysaccharide chitin and chitosan fibers, unlike higher plants, which is made up of cellulose and pectin. Chitin and chitosan inhibits the absorption of cholesterol and at the same time speeds up the metabolism.

Human beings become more vulnerable to various diseases as age advances. Ageing-related diseases and disorders include hypercholesterolemia, obesity, hypertension and hyperglycemia. Now a days, oyster mushroom are recognized as important food for their significant role in human health, nutrition and diseases. The use of mushroom nutrition as part of nutritional management to enhance the body's immune function is considered as standard practice in Japan, China and in other Asian cultures (Konno, 2003).

SUMMERY

It has been shown that a wide variety of agricultural (by-) products, weeds and wastes can be successfully used to produce food, feed, enzyme and medicinal compounds and to degrade and detoxify wastes. Due to an increasingly negative human impact on the environment, these techniques, together with others, constitute a very important tool for converting abundant quantities of waste materials, which often cause environmental pollution, into food and valuable compounds. These and many other materials have been successfully used for oyster mushroom production.

Oyster mushrooms can help our body by various ways. *Pleurotus* spp. have been used by human cultures all over the world for their nutritional value, medicinal properties and other beneficial effects. Oyster mushrooms are a good source of dietary fiber and other valuable nutrients. They also contain a number of biologically active compounds with therapeutic activities. Oyster mushrooms modulate the immune system, inhibit tumour growth and inflammation, have hypoglycaemic and antithrombotic activities, lower blood lipid concentrations, prevent high blood pressure and atherosclerosis, and have antimicrobial and other activities. As for cancer, research shows a possible anti-tumor effect from polysaccharides in oyster. A polysaccharide is a complex carbohydrate made up of smaller sugar molecules. Specific polysaccharides, known as ß-D-glucans, are suspected to stimulate the immune system to fight cancer. The ß -D-glucan isolated from oyster mushrooms is called pleuran. Studies are ongoing into the effects of pleuran for cancer treatment. However, the biochemical mechanisms of these therapeutic activities still remain largely unknown.

In addition to helping our body, oyster mushrooms can help our environment as well. The oyster mushroom is a saprotroph, meaning it feeds on dead and decaying matter (mainly wood) so the most fascinating use of these mushrooms is their growing role in mycorestoration. Mycorestoration is the process of using mushrooms to decrease pollution levels in a given area. The mycelia of oyster mushroom kill and eat nematodes

(small roundworms) and bacteria, making them one of the few carnivorous mushrooms. Oyster mycelium is ravenous. It will eats through wood, paper, coffee grounds, and even petroleum products. These mushrooms are found on hardwoods. They secrete enzymes that break down the organic bonds in wood into smaller molecules. The carbon-hydrogen bonds in wood are similar to those found in oil and pesticides. Thus due to their love of wood, oyster are also efficient in breaking down the organic bonds in toxic chemicals. In addition to breaking down the organic bonds in oil, oyster mushrooms are also powerful absorbers of mercury. Their mycelium channels mercury from the ground up into the mushroom itself. Once the mushroom is picked and destroyed, the mercury is removed from the environment. Thus it is not a dream to remove toxic heavy metals like mercury from our soil and water by cultivating mushrooms.

CONCLUSION

Considering the above facts it is of no doubt that oyster mushrooms are a crop of high nutritional and medicinal values which can be cultivated in our densely populated country without obstructing the limited fertile agricultural land. If due attention is paid by our government, its cultivation will be very much cost effective and may be one of our ideal food to combat the protein energy malnutrition very much prevalent in our country. Mushroom can prevent, control and even can cure many life threatening diseases. In this regard mushrooms could open new possibilities for treatment. Moreover, mushroom trading can help solving the unemployment problem of our country and reduce the import cost of medicine thereby can play an important role in our economy.

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