

EFFICIENCY OF OYSTER MUSHROOM (*PLEUROTUS COLUMBINUS* - P 8) USING DATE PALM LEAVES WITH COMBINATION OF WHEAT STRAW AND COTTON WASTE FOR ITS YIELD IMPROVEMENT

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Abstract

The prospects of cultivating Oyster mushroom (*Pleurotus columbinus*-P8) on date palm leaves with a combination of different agricultural waste, the supplementary material wheat bran, corn meal and gypsum were used at the rate of 5% of substrate dry weight in all treatments. Data were recorded from spawn running, pinhead formation, and growth of fruiting body, total number of fruiting bodies, yield and biological efficiency. Results revealed that treatment T4 (date palm leaves 25% + wheat straw and cotton waste 75%) took minimum days for spawn running, pinhead formation, comparative growth of fruiting bodies and produce a maximum number of fruiting bodies. While treatment T1 (date palm leaves 100%) gave maximum days for spawn running, pinhead formation, comparative growth of fruiting bodies and produce a minimum number of fruiting bodies. The yield production and biological efficiency of Oyster mushroom was found to be higher in treatment T4, (date palm leaves 25% + 75% (wheat straw + cotton waste) gave 403 g and 80.6% respectively. While yield production and biological efficiency oyster mushroom was found lowest in treatment T1 (date palm leaves=100% gave (185.75) g and (37.15) % respectively. Therefore, the technology of mushroom agriculture enables us to acquire substrate materials at very low cost and further led to conserve our environment through efficient bioconversion of wastes with sustainable food security.

Key words: Agricultural waste, Supplementary material, Spawn running, Pinhead formation, Biological efficiency.

Introduction

Mushroom (*Pleurotus columbinus*-P8) belongs to Phylum Basidiomycota, Class: Basidiomycetes and Family: Agaricaceae of the Kingdom Fungi. Oyster mushroom as a food is the third major cultivated mushroom. Fungus is used as food for human consumption, it is a rich source of protein contents ranges (1.6–2.5 %) and vitamin C and B complex (Randive, 2012). In developing countries when meat sources are limited than mushrooms is a good dietary source of protein (Eswaran & Ramabadran, 2000). It can play a vital portion in inspiring human foods with protein substances reaching from (25-40%) when dry and 3-7% when fresh in the shortage of meat sources or when meat sources are inadequate. Protein contents are higher than the mushroom in most vegetables and fruits with the exclusion of peas and beans. In developing countries mushroom is an outstanding source of nourishment for diet due their texture, dietary value, taste and extraordinary output per unit area (Eswaran & Ramabadran, 2000). In the world wide *Pleurotus* species are the main source of edible mushroom, so it is necessary to select a suitable substrate for the production of oyster mushroom. Four different species of oyster mushroom was cultivated on saw dust and rice bran substrate for its mycelial growth and yield efficiency. *P. salmoneostramineus* and *P. ostreatus* grey gave highest mycelial growth which is 1.29 mm/day and 1.17 mm/day respectively. Similarly *P. salmoneostramineus* and *P. ostreatus* grey gave highest growth which was 12.90 cm/day and 11.74 cm/day respectively in 30 days old bags (Owaid *et al.*, 2015). Oyster mushroom (*P. ostreatus*) is one of the commonly like eatable fungus cultured artificially in South East Asian nations in the subtropical and moderate region. It is well known all over the world due to their brilliant taste and

flavor. Mushrooms are the home for usual power and virility and are used in making of many sub continental dishes and have therapeutic properties like anticholesteral, anticancerous and antitumorous. It is normally cultivated all over the world, particularly in South East, Europe, Africa and Asia. On dry mass base, the genus is categorized by its abundant protein content, i.e. 30-40 % (Sharma & Madan 1993). For the cultivation of oyster mushrooms, numerous agricultural waste materials are used as a substrate like rice husk, cotton waste, tea leaves, banana leaves, wheat straw, kikar saw dust and rice straw etc. (Thomas *et al.*, 1998). Tea waste was used as substrate for the cultivation of oyster mushroom. This waste contains re-useable energy and nutrients which pollute the environment (Yang *et al.*, 2016). In Pakistan all types of edible mushroom are excellently grown due to good climatic conditions. In plain areas of Sindh, Baluchistan, K.P.K and Punjab, substrate is available at low cost for the cultivation of mushroom and more than 100 countries of the world, mushrooms are cultivated with an average production of (12) million tons (Suman and Sharma, 2007). In Pakistan during rainy season fungi are observed on the humid with plenty of humus and dunghill. If suitable source of food and moisture is available for growth, fungus can be easily grown in local condition (Ibekwe *et al.*, 2008). The supplements improve the nutritional rank of cotton waste substrate which resulted in enhanced mushroom growth (Khan *et al.*, 2017). Cotton is an important crop of Asia In Pakistan the cultivation of mushroom has been started by the National Logistic Cell (NLC) in the areas of Swat and Islamabad with an annual production ability of 48 tons. To earn foreign exchange about 80% of the total production is exported (Latif & Shinwari, 2006; Alam & Raza, 2001; Akhtar, 1992). Date palm (*Phoenix dactylifera*) is a vital ancient monocotyledonous plant. It

composed of trunk, leaves, seeds, and fruits. Date palm leaves are (100–250) cm long and will reach up to (4–7) long, (Al-Gboori & Krepl, 2010). Date palm components are obligatory by-products of date production and their main use is focused to boost soil organic content, animal feed, and manifested feedstock resembling polysaccharide and hemicelluloses. A high proportion of polysaccharide and polymer are found in the numerous components of the date palms, bases and midribs, leaflets, fruit stalks. These components are often used as a substrate to grow alimentary product with low price resembling an oyster mushroom. Moreover, Date palm leaves contain minerals resembling N, P, K, Ca, Mg, and Iron reported by (Abo Hassan & Bacha, 1982). Because of the well-known capability to convert food proteins, Oyster mushrooms are the easiest and least expensive commercial mushrooms to grow. Therefore, the study was carried out to investigate the influence of date palm leaves with combined effect of other cellulosic waste material for yield production of *P. columbinus*-P8.

Materials and Methods

Collection and preparation of substrate: For the preparation of substrate date palm leaves were collected from the Date Palm Research Institute Jhang, Punjab, Pakistan and agricultural waste materials (wheat straw and cotton waste) were collected from the Department of Agronomy and Local market of Faisalabad, Pakistan respectively. The date palm leaves were sun dried and sliced into small pieces (2–3 cm long) with the help of the chopper. The small pieces of date palm leaves and agricultural waste material (wheat straw and cotton waste) were soaked in water for about (18–24 hours) and excess of moisture was drained off by placing the material on newspapers. After that soaked material was placed on the cement floor to get desired moisture (70%) of the substrate. Gypsum and supplements of (corn meal and wheat bran) were added to the mixture at the rate of 5% of the substrate dry weight (Alananbeh *et al.*, 2014). Before filling the bags fermentation of substrate was done in 5 days by covering with polythene sheets. After fermentation mixing of all material were done as per treatment ratios.

Filling and sterilization of bags: Heat tolerant polypropylene bags (8"×12") were filled with fermented substrate. A total 500g of substrate was filled in each polypropylene bag and their mouths of the bags were sealed with the help of rubber bands. Sterilization of bags containing fermented substrate was done in the country style (drum) autoclave for 2 hours. After sterilization bags were kept at room temperature for cooling for 24 hours.

Inoculation, incubation and culture conditions: Prepared spawn of oyster mushroom was taken from the Mushroom Laboratory Institute of Horticultural Sciences, University of Agriculture Faisalabad. For inoculation of sterilized bags 15 g of spawn was used per bag for all above mentioned treatments and replications. These inoculated bags were incubated in growth room at 20–25°C under 80–90% relative humidity with complete darkness until the substrate was completely colonized

with mycelium. After the growth of mycelia holes were made into bags by using clean knife.

Harvesting parameters: Mushrooms were harvested from the substrate when the caps got fully mature and before the fruiting bodies start to curl up. The clusters of the mushrooms were weighed, and several parameters were evaluated: the length of each phase of the fungus production cycle (first and second flush), earliness (the time elapsed between the day of inoculation and the day of the first harvest), fruiting body number per flush and in total, the average weight of individual fruiting body per flush and in total, the average yield for each treatment per flush and in total, and biological efficiency.

$$\text{B.E (\%)} = \frac{\text{Fresh weight of mushroom harvested}}{\text{Substrate dry matter}} \times 100$$

Statistical analysis: The experiment was laid out in completely randomized design and the data recorded was statistically analysed to determine the significance of the experiment. The Least Significant Difference test was applied to the data at probability level of 5% that gave a practical and comprehensive comparison of the treatment for their effectiveness.

Results and Discussions:

Accomplishment of spawn running in days: The studies on the completion of 25%, 50%, 75% and 100% mycelium growth of Oyster mushroom on substrates showed in Fig. (1) that treatment T4=(date palm leaves 25% + agricultural waste material 75% (wheat straw + cotton waste) took minimum days (6.5, 13.0, 17.5 and 22.5) and treatment T1 (date palm leaves 100%) took maximum days (16.0, 21.0, 28.5 and 35.5) for the accomplishment of 25%, 50%, 75% and 100% mycelium growth for Oyster mushroom while, the remaining treatments took different number of days such as treatment T2=(date palm leaves 75% + 25% (wheat straw + cotton waste)took (14.5, 18.5, 26.5 and 32.5) days, treatment T₃=date palm leaves 50% + agricultural waste material 50% (wheat straw + cotton waste) took (9.5, 15.5, 20.0 and 26.0) days and treatment T5=(Agricultural waste material 100% (wheat straw + cotton waste) took (12.0, 17.0, 24.0 and 29.0) days for 25%, 50%, 75% and 100% accomplishment of spawn running. By performing the analysis of variance (ANOVA) indicate that treatment T4=(date palm leaves 25% + agricultural waste material 75% (wheat straw + cotton waste) differ significantly ($p < 0.05$) for accomplishment of 25%, 50%, 75% and 100% mycelium growth of oyster mushroom (*P. columbinus*-P8) as compared to all other treatments. Presence of cellulose in the substrate and nature of temperature affect the process of spawn running (fast or slow). The substrates having higher amount of cellulosic and lignocellulosic element are suitable for the fast growth of mushroom (Quimio, 1976). Accomplishment of spawn running is affected by the different chemical composition of the substrate (Jonathan *et al.*, 2012). The mycelium growth rate is also affected by the presence of different kind of polyphenolic substances in these substrates (Zadrazil, 1982).

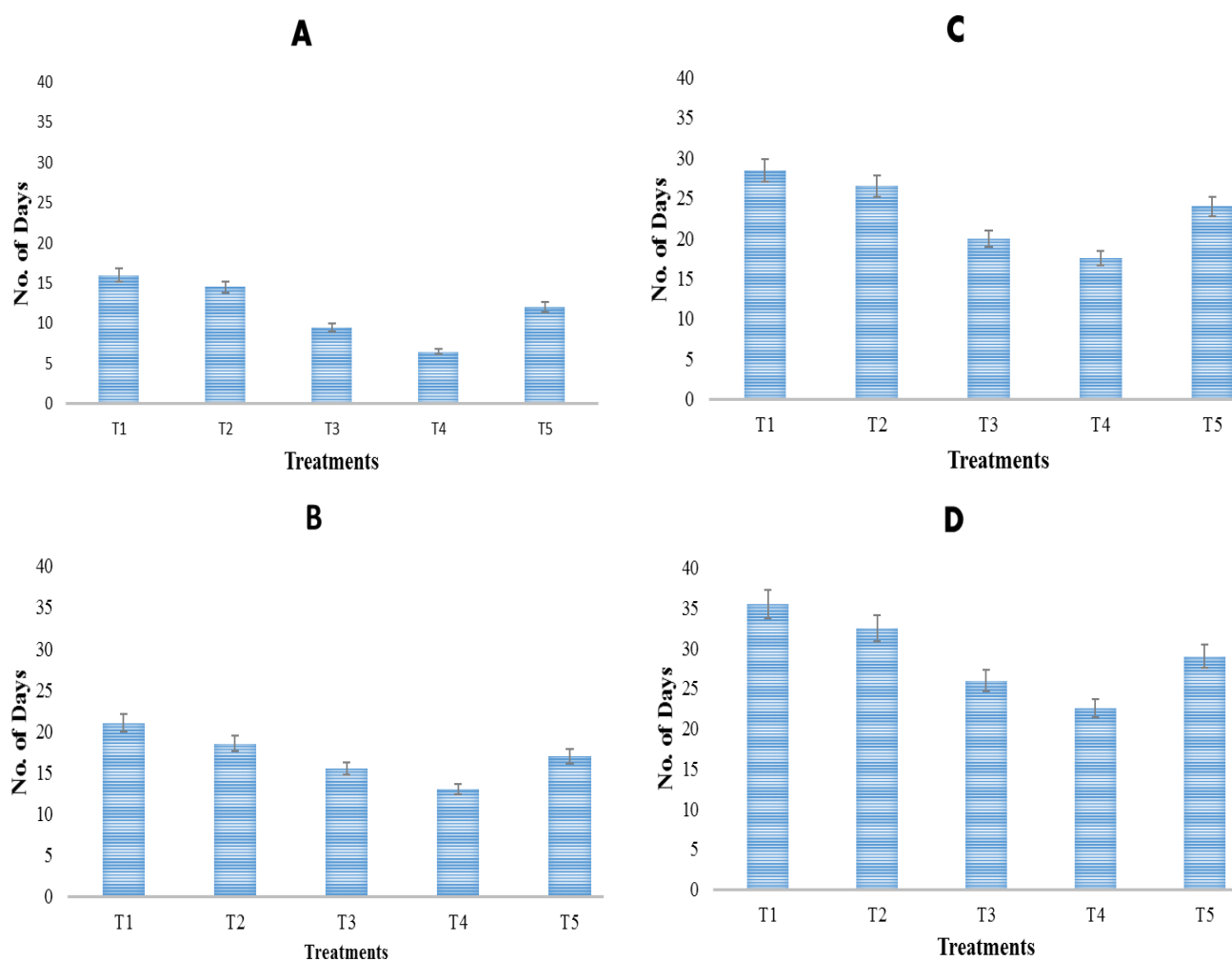


Fig. 1. Accomplishment of spawn running in days (A;25%, B;50%, C;75% and D;100%).

Comparative growth of pinhead formation in days:

The data related to the formation of a pinhead in number of days that treatment T4= date palm leaves 25% + 75% (wheat straw + cotton waste) took minimum days (5.50) and treatment T1 = date palm leaves 100% took maximum days (15.00) for the development of pinhead for oyster mushroom (Fig. 2). While the remaining treatments took a different number of days such as treatment T2 took (12.50) days, treatment T3= date palm leaves 50% + 50% (wheat straw + cotton waste) took (8.00) days and treatment T5 = (100% (wheat straw + cotton waste) took (9.50) days for the development of pinhead on these substrates after the bags fully covered with mycelium and become white. By performing the analysis of variance (ANOVA) Treatment T4 differed significantly ($p < 0.05$) for the development of pinhead formation of oyster mushroom (*P. columbinus* - P8) with relate to all other treatments. Pinhead commencement is reliant on mycelium growth as they initiate when the substrate is fully filled with fungal mycelium. Cultivation of *Pleurotus* spp. was done on different lignocellulose and cellulose substrates, to spawn accomplishment and pinhead formation different temperature 16-20°C and 20-22°C respectively were observed (Shah *et al.*, 2004).

Comparative growth of fruiting body formation in days:

The data related to maturation of fruiting bodies after the formation of pinhead is shown in Fig. (3). Treatment T4 took minimum days (5.0) and treatment T1 took maximum days (13.0) for the development of fruiting bodies. While the remaining treatments T2 took (11.5), treatment T3 took (7.0) and treatment T5 took (9.5) number of days for the maturation of fruiting bodies after the formation of pinheads. By performing the analysis of variance (ANOVA) it is stated that treatment T4 differed significantly ($p < 0.05$) for maturation of fruiting bodies of oyster mushroom (*P. columbinus*-P8) with relate to all other treatments. The difference in late maturation of fruiting body development is maybe due to ecological situation or might be the excessive makeup of (*Pleurotus* spp.) use in this study. The outcomes are in agreement with the findings of previous workers (Baysal *et al.*, 2003; Tisdale *et al.*, 2006). Indol Acetic Acid (IAA) enhance the total number of fruiting bodies of mushroom described by (Poppe *et al.*, 2000).

Total number of fruiting bodies: The results indicate the total number of fruiting bodies after the formation of pinhead on these substrates that treatment T4 has maximum number of fruiting bodies (36.5) and treatment T1 has minimum (9.5) number of fruiting

bodies (Fig. 4). While the remaining treatments T2 produced (15.5) number of fruiting bodies, treatment T₃ produced (27.0) number of fruiting bodies and treatment T5 has (20.0) number of fruiting bodies after the formation of pinheads. By performing the analysis of variance (ANOVA) it is stated that treatment T4 differed significantly ($p < 0.05$) for total number of fruiting bodies of oyster mushroom (*P. columbinus*-P8) with relate to all other treatments. Presence of substances such as Glucose, fructose and cellulose and environmental factor light, temperature and humidity affect the total numbers of fruiting bodies on these substrates (Kalita *et al.*, 1997).

Number of flushes and yield on each flush (g):

Harvesting of mushroom was done at the maturity of each flush; three flushes of mushroom were taken from each treatment. Weight of harvested mushroom was taken in grams shown in Fig. (5) that treatment T4 gave the maximum yield production (165.75, 133.0 and 104.25 g) and treatment T1 gave minimum yield production (86.75, 63.0 and 36.0 g) in case of each flush on substrates. While remaining treatments T2 gave (103.50, 83.75 and 56.0 g) yield, treatment T₃ gave (143.50, 114.75 and 83.75 g) yield and treatment T5 gave (126.75, 97.50 and 73.50 g) yield on these substrates. As per analysis of variance (ANOVA) it is stated that treatment T4 differ significantly ($p < 0.05$) for yield production of oyster mushroom (*P. columbinus*-P8) with relate to all other treatments. The outcomes are correlated with (Mane *et al.*, 2007; Islam *et al.*, 2009; Obodai *et al.*, 2003; Omoanghe *et al.*, 2009; Olfat & Payvast, 2008) suggest that maximum yield of mushroom depends upon the presence of xylanase enzymes in the substrates.

Total yield of Oyster mushroom (g): Studies about the total weight of oyster mushroom is shown in Fig. (6) that treatment T4 gave the maximum yield production (403.0 g) and treatment T1 gave minimum yield production (185.75 g) on these substrates. While remaining treatments T2 gave (243.25 g) yield, treatment T₃ gave (342.0 g) yield and treatment T5 gave (297.75 g) yield of oyster mushroom on these substrates. By performing the analysis of variance (ANOVA) it is stated that treatment T4 differ significantly ($p < 0.05$) for yield production of oyster mushroom (*P. columbinus*-P8) with relate to all other treatments.

Biological efficiency of Oyster mushroom: Data taken about the biological efficiency of Oyster mushroom showed that treatment T4 had maximum biological efficiency (80.60%) and treatment T1 gave minimum biological efficiency (37.15%) on these substrates while remaining treatments T2 gave (48.65%) biological efficiency, treatment T₃ had (68.40%) biological efficiency and treatment T5 had (59.55%) biological efficiency of oyster mushroom on these substrates (Fig. 7). By performing the analysis of variance (ANOVA) it is stated that treatment T4 differed significantly ($p < 0.05$) for biological efficiency of oyster mushroom (*P. columbinus*-P8) with relate to all other treatments.

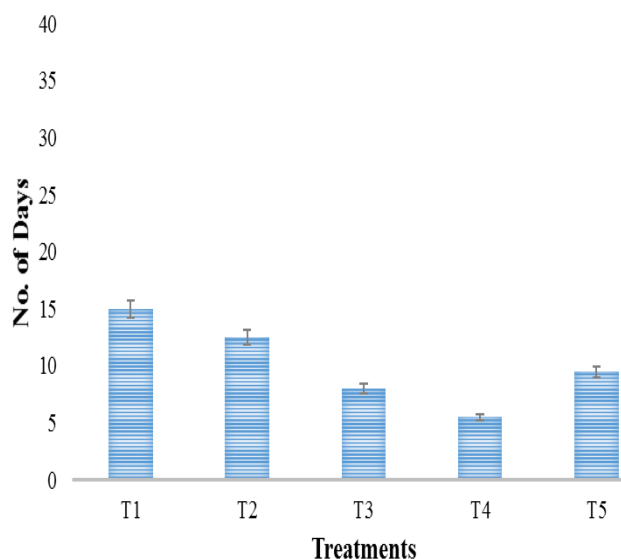


Fig. 2. Comparative growth of pinhead formation in days.

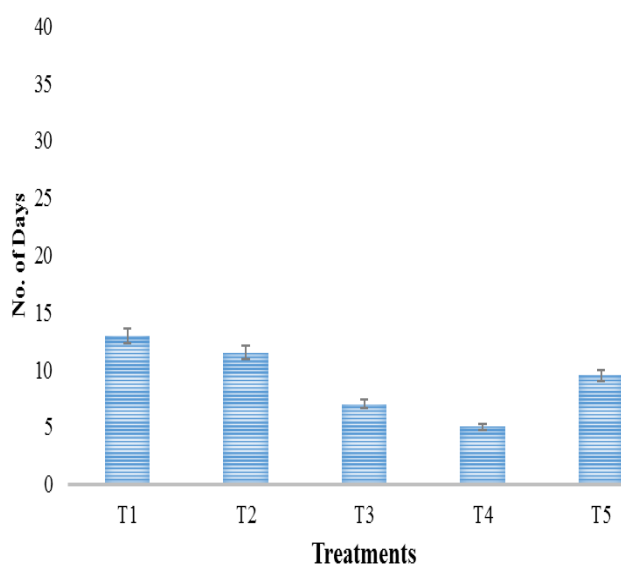


Fig. 3. Comparative growth of fruiting body formation in days.

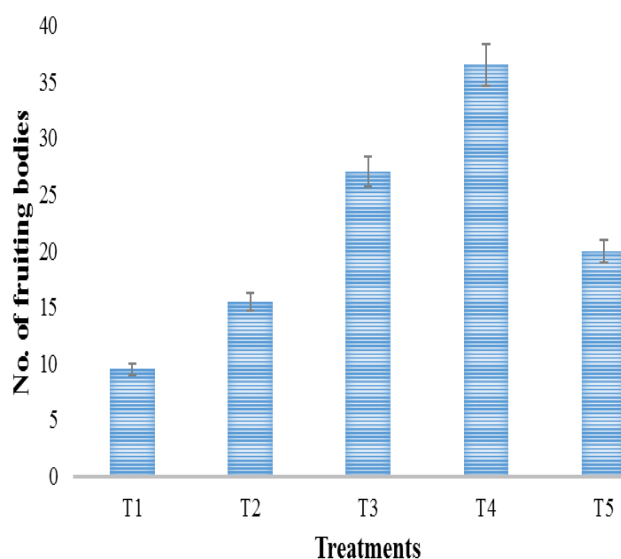


Fig. 4. Total number of fruiting bodies.

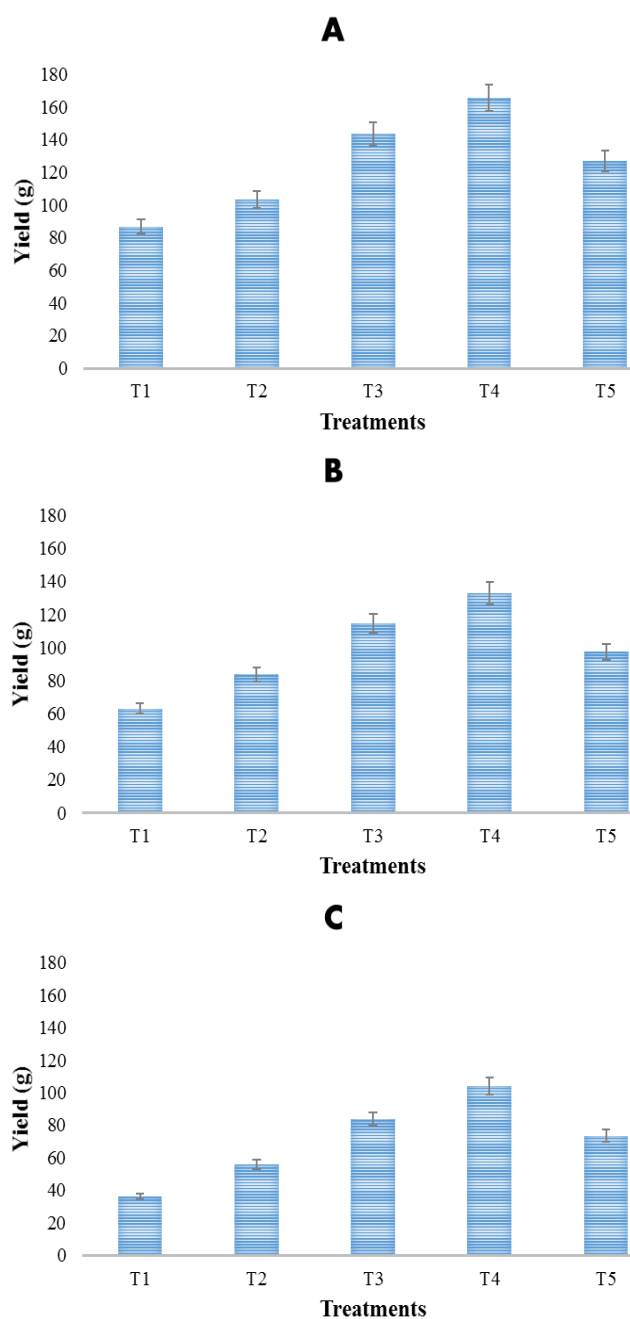


Fig. 5. Number of flushes and yield on each flush in grams (A;1st Flush, B;2nd Flush and C;3rd Flush).

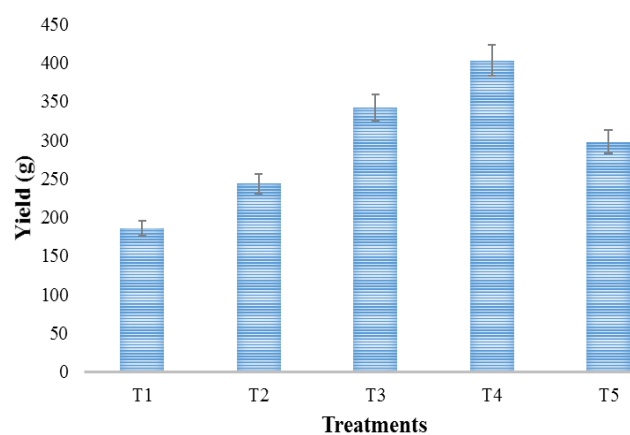


Fig. 6. Total yield of Oyster mushroom in grams.

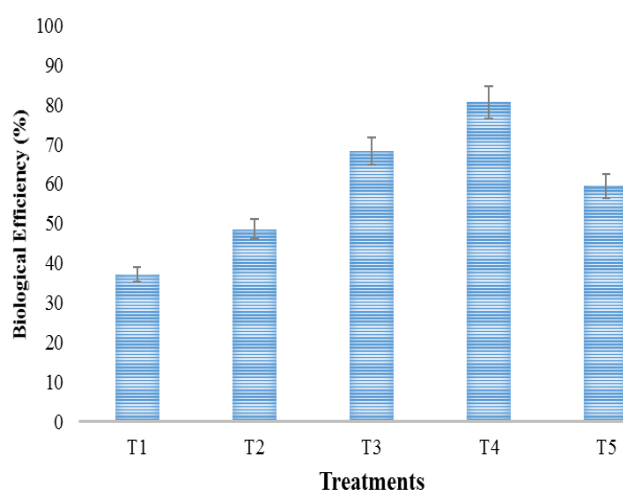


Fig. 7. Biological efficiency of Oyster mushroom.

References

Abo Hassan, A.A. and M.A. Bacha. 1982. Mineral composition of the foliage of four Saudi Arabian date palm cultivars. *J. Coll. Agric. King Saud Univ.*, 4: 35-42.

Akhtar, T. 1992. *Effect of different doses of compost on the yield of Agaricus biosporus*. M.Sc. thesis. Dept. of Bot. Uni. of Peshawar, Pakistan.

Alam, S.M. and M.S. Raza. 2001. Importance of Mushrooms. Industry and Economy. NIA, Tandojam, Pakistan.

Alanbeh, K.M., N.A. Bouqellah and N.S. Al-Kaff. 2014. Cultivation of oyster mushroom *Pleurotus osteratus* on date palm leaves mixed with other agro-wastes in Saudi Arabia. *Saudi J. Biol. Sci.*, 21: 616-625.

Al-Ghoori, B. and V. Krepl. 2010. Importance of date palms as a source of nutrition. *Agric. Trop. Subtrop.*, 43: 348-351.

Baysal, E., H. Peker, M.K. Yalinkilic and A. Temiz. 2003. Cultivation of oyster mushroom on waste paper with some added supplementary materials. *Biores. Technol.*, 89: 95-97.

Eswaran, A. and R. Ramabadran. 2000. Studies on some physiological, cultural and postharvest aspects of oyster mushroom (*Pleurotus ostreatus*). *Tropical Agri. Res.*, 12: 360-374.

Ibekwe, V.I., P.I. Azubuike., E.U. Ezeji and E.C. Chinakwe. 2008. Effects of nutrient sources and environmental factors on the cultivation and yield of Oyster Mushroom (*Pleurotus ostreatus*). *Pak. J. Nutr.*, 7: 349-351.

Islam, M.Z., M.H. Rahmani and F. Hafiz. 2009. Cultivation of Oyster mushroom (*Pleurotus flabellatus*) on different substrates. *Int. J. Sustain. Crop Prod.*, 4: 45-48.

Jonathan S.G., C.B. Okon, A.O. Oyelakin and Olurantioo. 2012. Nutritional values of oyster mushroom (*Pleurotus ostreatus*) cultivated on different agricultural wastes. *Nature & Sci.*, 10: 186-191.

Kalita, M.K., Y. Rathiah and K.N. Bhagabati. 1997. Effect of some agro-wastes a substrate for oyster mushroom cultivation in Assam. *Ind. J. Hill Farming*, 19: 109-110.

Khan, N.A., A. Amjad, R. Binyamin, A. Rehman and O.A. Hafeez. 2017. Role of various supplementary materials with cotton waste substrate for production of *Pleurotus ostreatus* on Oyster mushroom. *Pak. J. Bot.*, 49(5): 1911-1915.

Latif, A. and Z.K. Shinwari. 2006. Sustainable market development for non-timber forest products in Pakistan. Uni. of Sci. and Technol. Kohat, Pakistan.

Mane, V.P., S.S. Patil, A.A. Syed and M.M.V. Baig. 2007. Bioconversion of low quality lignocellulosic agricultural waste into edible protein by *Pleurotus sajor caju*. *Singer. J. Zhejiang. Uni. Sci.*, B. 8: 745-751.

- Obodai, M., J.C. Okine and K.A. Vowotor. 2003. Comparative study on the growth and yield of *Pleurotus ostreatus* mushroom on different lignocellulosic byproducts. *J. Indi. Microbiol. Biotechnol.*, 30: 146-149.
- Olfati, J.A. and G. payvast. 2008. Lawn Clippings for Cultivation of Oyster Mushroom. *Int. J. Veg. Sci.*, 14: 98-103.
- Omoanghe, S. Ishikhuemhen and N.A. Mikiashvilli. 2009. Lignocellulolytic enzyme activity, substrate utilization and mushroom yield by *Pleurotus ostreatus* cultivation on substrate containing anaerobic digester solids. *J. Ind. Microbiol. Biotechnol.*, 36: 1353-1362.
- Owaid, M.N., S.S. Al-Saeedi, V. Sabaratnam, I.A.A. Al-Assaffii and J. Raman. 2015. Growth performance and cultivation of four oyster mushroom species on saw dust and rice bran substrates. *J. Adv. Biotechnol.*, 4(3): 424-429.
- Poppe, J., H.K. Goering and V. Soest. 2000. Use of agricultural waste materials in the cultivation of mushrooms. *Mushroom Sci.*, 15: 3-23.
- Quimio, T.H. 1976. Cultivation ganoderma the "Pleurotus" mushroom. *Newsletter. Tropics*, 6: 128-130.
- Randive, S.D. 2012. Cultivation and study of growth of Oyster mushroom on different agricultural waste substrate and its nutrient analysis. *Adv. Appl. Sci. Res.*, 3: 1938-1949.
- Shah, Z.A., M. Ashraf and M.Ch. Ishtiaq. 2004. Comparative study on cultivation and yield performance of Oyster Mushroom (*Pleurotus ostreatus*) on different substrates (wheat straw, leaves, saw dust). *Pak. J. Nutr.*, 3: 158-160.
- Sharma, S. and M. Madan. 1993. Microbial protein from leguminous and non-leguminous substrates. *Acta Biotechnol.*, 13: 131-139.
- Suman and Sharma. 2007. Mushrooms cultivation in India, Daya publishing house, Delhi, 1100: 18-22.
- Thomas, G.V., S.R. Prabhu, M.Z. Reeny and B.M. Bopaiah. 1998. Evaluation of lignocellulosic biomass from cocoonut palm as substrate for cultivation of *Pleurotus sajor caju*. *World J. Microb. Biotechnol.*, 14: 879-882.
- Tisdale, W., P.S. Patil and A. Jadhav. 2006. Oyster mushroom cultivation on different woods of trees. *Mushr. Biol. Mushr. Prod.*, 3: 11-16.
- Yang, D., J. Liang, Y. Wang, F. Sun, H. Tao, Q. Xu, L. Zhang, Z. Zhang, C.T. Ho and X. Wan. 2016. Tea waste an effective and economic substrate for oyster mushroom cultivation. *J. Sci. Food and Agriculture*, 96(2): 680-684.
- Zadrazil, F. 1982. Conversion of different plant waste in to feed by Basidiomycetes. *Champagon*, 135: 25-32.

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