Biodegradation of Lignocellulosic Wastes by Cultivation of Mushrooms as Nutrient Source

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Abstract: India being an agriculture based country produces a large amount of lignocellulosic wastes which is either left to rot or is burned in the fields causing several environmental problems. Several mushrooms of high nutritional values are known to grow in these lignocellulosic wastes. Cultivation of such mushrooms using lignocellulosic wastes will not only solve the problem of lignocellulose biodegradation, but also act as a source of nutrition in the rural areas where malnutrition is prevalent.

Keywords: lignocellulose, mushroom cultivation, nutritional, lignocellulosic waste

1. Introduction

Lignocellulose is an organic matter which is renewable in nature and is structural component of all the plants, due to presence of lignin it becomes recalcitrant to bio-degradation. Different industries such as forestry, paper, agriculture, food and paper pulp produce a large amount of lignocellulosic waste each year [1]. These materials with high potential value for the production of many valuable substances are treated as waste in many developing countries and still are either left out to rot or incinerated causing serious environmental problems [2]. There have been many fungi identified which are known to degrade lignocellulosic waste, but all of them are not rendered fit to eat. Edible mushrooms are the fruiting bodies of the fungi, generally of the Phylum Basidiomycota. The extracellular enzyme system of different rot fungi are capable of degrading the lignocellulosic material and use them as their substrate to grow and produce fruiting bodies. These mushrooms are known to have high nutritional value. In developing countries like India where 19.8 million children below the age of 6 years are undernourished (ICDS 2015) and only 9.6% children between 6 months to 2 years receive an adequate diet (NFHS 4, 2015), cultivating the large quantity of mushrooms on economic and readily available lignocellulosic substrates from agro industrial wastes can help the country in fighting against the malnutrition and also in safe biodegradation of the lignocellulosic waste. This review focuses on different types of edible mushrooms and the lignocellulosic wastes they can grow upon.

2. History

Mycophagy has been long practiced in human civilization. The most primitive evidence found is 13,000 year old in Ötzi, the mummy which was buried with two species of edible mushrooms. The first mushroom found to be growing in the agricultural compost was A. bisporus, it was found growing in the melon compost. Earlier it was grown in open fields but later on its cultivation was moved to underground caves as it had more suitable atmospheric conditions. Mushroom cultivation in USA was introduced in 1865, in 17th century in Europe and in 19th century in Netherlands. Japan and China are considered to be the first nations who exploited the medicinal uses of the mushrooms.

3. Medicinal and Nutritional Properties of the Mushrooms

Since a long times the mushrooms have been known to contain medicinally beneficial compounds and nutritious to health. Ledodes were grown in china just for their medicinal uses with the AIDS drugs [3]. P.ostreatus contains high values of carbohydrates, proteins, vitamins and minerals [4]. Many species of the mushrooms have been proven to be a great source of medicinally useful compounds to treat severe ailments such as, Ledodes is known to lower blood cholesterol levels and blood pressure. Tremella fuciformis spore extracts perform antilipemic activity, Grifola frondosa is known to lower blood pressure without changing plasma HDL levels and Trametes sp. is known for its antihyperlipemic and antiarrhythmic effects [5]. The medicinal properties of some important culinary mushrooms are given in table 1.

Table 1: Medicinal properties of some mushroom species

| Sr no. | Name of species | Medicinal importance | Reference |
|--------|-----------------|----------------------|-----------|
| 01     | Auricularia polytrechia | Antioxidant activity | Mau et al. 2000 |
|        |                  | Immunomodulatory     | Sheu et al. 2004 |
|        |                  | Antitumor            | Yu et al. 2004; Song and Du 2012 |
|        |                  | Anti-dementia activity | Bennet et al. 2013 |
|        |                  | Attenuation of inflammatory response and oxidative stress | Chiu et al. 2014 |
| 02     | Pleurotous ostreatus | Extracts lower cholesterol | Khatun et al. 2007 |
|        |                  | Antinociceptive      | Jayakumar et al. 2009 |
|        |                  | Antitumor            | Vasisdewa et al. 2008 |
|        |                  | Antioxidant and immunological activities | Sarangi et al. 2006 |
| 03     | Pleurotous florida | Antioxidant and antitumor properties | Josan J.K. 2000 |

4. Sources of lignocellulosic waste

States and districts in India have access to a large no. of lignocellulosic wastes which are generated from farms,
forests, processing industries and kitchen wastes. The major sources of lignocellulosic biomass are as below.

4.1 Agricultural residue

The agriculture is a major source of income for the rural livelihood and in the huge production of the crops large amount of agricultural residues are also generated. These residues can be classified into two types, primary and secondary. The residues that are left in the fields such as straw comes under primary residue and the products such as husk which are obtained after the processing of the crop are known as the secondary residue. All the agro lignocellulosic materials such produces have high biomass potential in range of 15000 – 1.5 lakh (kilo tonnes/year) [6],[7].

4.2 Industrial

Lignocellulosic by-products from many industries such as paper, pulp, breweries etc. are released and causes various types of pollution. For example, the coffee production worldwide, for the production of per ton fresh coffee, produces 0.5 tons of coffee pulp and 0.18 ton husk per year [8], and also per year six million tons of spent coffee grounds are produced [9]. Caffeine present in the SCG is eco-toxic to a considerable limit and this presence of caffeine also presents hindrance to its many applications [10]. A range of filamentous fungi have been recorded to degrade the caffeine [11], but these have been only studied for the industrial degradation of caffeine containing wastes [12],[13].

4.3 Food and kitchen wastes

Left over foods, vegetables peels, fruit scraps etc. usually remain untreated and left to rot in open causing many environmental and health problems.

4.4 Forest waste

Forests are found in abundance throughout the world at all times of the year irrespective of season and food habits of the people. Sawdust, leaves, logs, twigs, barks are among the lignocellulosic materials that can be obtained from the forests throughout the year.

5. Composition of lignocellulosic residue

Lignocellulose is an organic matter and acts as structural component of plants. Cellulose, hemicellulose and lignin are the prime constituent of the lignocellulosic materials. Cellulose and hemicellulose are sugar derivative macromolecules; whereas lignin are the aromatic polymers made from the phynylpropanoid precursors. The composition and ratio of these molecules may vary with different plants, [14]compiled a list of the same.(Table 2).

6. Bioconversion of lignocellulose

Major components of the lignocellulosic materials are cellulose, hemicellulose and lignin with varying composition in different materials [15]. It is difficult to dissolve lignin without destroying its structure, hence it becomes tedious to ascertain its exact chemical structure. Normally lignin consists of three aromatic alcohols but the lignin found in dicots and grasses also contains phenolic acids esterified to alcohol groups. The lignin present in lignocellulose is also found to be attached with cellulose and hemicellulose covering them and creating a impermeable physical barrier. Due to these physical attributes it becomes recalcitrant to degradation [16]. Several chemical methods such as alkaline hydrolysis [17] and acid hydrolysis [18] have been proposed for the degradation of the lignocellulose, but, these methods produce undesirable

| Substrate               | Hemicellulose | Lignin | Cellulose | References                  |
|-------------------------|---------------|--------|-----------|-----------------------------|
| Hardwood stem           | 24-40         | 18-25  | 40-45     | Howard et al. (2003), malherbe and cloete (2002). |
| Coastal Bermuda grass   | 35.7          | 6.4    | 25        | Howard et al. (2003)        |
| Sorted refuse           | 20            | 20     | 60        | Howard et al. (2003)        |
| Corn cobs               | 35            | 15     | 45        | Howard et al. (2003), prassad et al. (2007), mckendry (2002). |
| Grasses                 | 35-50         | Oct-30 | 25-40     | Howard et al. (2003), malherbe and cloete (2002). |
| Paper                   | 0             | 0-15   | 85-99     | Howard et al. (2003)        |
| Rice straw              | 24            | 18     | 32.1      | Howard et al. (2003), prassad et al. (2007), mckendry (2002). |
| Nut shells              | 25-30         | 30-40  | 25-30     | Howard et al. (2003)        |
| Leaves                  | 80-85         | 0      | 15-20     | Howard et al. (2003)        |
| Cotton seed hairs       | May-20        | 0      | 80-95     | Howard et al. (2003)        |
| Newspaper               | 25-40         | 18-30  | 40-55     | Howard et al. (2003)        |
| Solid cattle manure     | 1.4-3.3       | 2.7-5.7| 1.6-4.7   | Howard et al. (2003)        |
| Softwood stem           | 25-35         | 25-30  | 45-50     | Howard et al. (2003), malherbe and cloete (2002). |
| Switch grass            | 31.4          | 12     | 45        | Howard et al. (2003)        |
| S32 rye grass (early leaf) | 15.8       | 2.7    | 21.3      | Howard et al. (2003)        |
| Orchard grass           | 40            | 4.7    | 32        | Howard et al. (2003)        |
| Wheat straw             | 26-30         | 16-21  | 29-35     | Rowell (1992), prassad et al. (2007), mckendry (2002). |
| Barley straw            | 24-29         | 14-15  | 31-34     | Rowell (1992).              |
| Oat straw               | 27-38         | 16-19  | 31-37     | Rowell (1992).              |
| Rye straw               | 27-30         | 16-19  | 33-35     | Rowell (1992), Stewart et al. (1997), Hon (2000). |
| Bamboo                  | 15-26         | 21-31  | 26-43     | Rowell (1992), reguant and rinaudo (2000), hon (2000). |
| Coffee pulp             | 46.3          | 18.8   | 35        | Pérez-diaz et al. (2005).   |
| Banana waste            | 14.8          | 14     | 13.2      | John et al. (2006).         |
byproducts and also the chemicals are hard to dispose hence the enzyme treatment is preferred over them. Further it has been shown that fungal extracellular enzymes such as cellulases are capable of converting lignocellulose into simpler compounds [19].

This lignocellulose degrading ability of the fungi can be attributed to their highly well-organized enzymatic system. There are two types of extracellular enzyme system, one which produces hydrolases for the degradation of polysaccharides and another one a unique extracellular and oxidative liginolytic system, which cleaves open phenyl rings and thus degrades lignin [20], a schematic representation of lignin degradation by white rot fungi was given [21] (fig 1).

Figure 1: Schematic representation of lignin degradation by white rot fungi.

7. Substrate preparation and supplements

In the inceptive period of mushroom cultivation a solid fermentation process is involved. Before bearing the fruiting body the mycelium grows in an aseptic controlled medium from spawns to colonize the whole substrate [22]. Depending on the requirement of the different species of mushrooms two methods using agricultural by-products for the preparation of substrates have been employed.

1). Composted substrates prepared by fermentation and pasteurization [23],[24]. This type of substrate is useful in the cultivation of Agaricus bisporous, P. sajor-caju and Pleurotus ostreatus[25],[26],[27].

2). Non-composted substrates that are usually mixture of various agricultural by-products used as main components of the substrate system and stem sterilized before the inoculation of the spawns. The species best suited for this method are, Auricularia sp[28], Pleurotus eryngii [29], Flammulina velutipes[30], Agrocybe aegerita [31], volvarella volvacea[32] and Lentimula edodes[27]. The C:N ratio is unique for each mushroom variety and when the substrate is supplemented with this ratio in adequate amount the highest yield in a short span of time can be achieved [33]. Apart from the commercial supplements many agricultural by-products are being used as nutritional supplements. When substrates having high concentration of alperujo were used for the cultivation of Flammulina velutipes, it was observed that the phytotoxicity of this by-product was reduced and the same time the mushroom showed good biological efficiency [34]. In Spain grapeseed meal, defatted pistachio and almond meal showed the same result for the cultivation of A. bisporous and P.ostreatus [35]. Several experiments as adding 25% of black bean, wheat bran and using soyabean, oat husk, and peanut shell with soya as supplement to P. ostreatus increased the protein of mushroom [36],[37]. Table 3 shows the various lignocellulosic materials used for different mushroom cultivation.

| Sr. no | Substrate composition | Mushroom variety | Reference |
|--------|-----------------------|------------------|-----------|
| 1      | Barley straw + sugarbeet pulp + rice bran | P.eryngii | Zadraji F. et al. 1992 |
| 2      | vegetable + paddy straw | Pleurotus | Ralph et al. 1994 |
| 3      | Soyabean straw + wheat straw | P. sajor-caju | Rani et al. 2008 |
| 4      | paddy straw + neem cake + black and red gram husk + cotton seed cake | Calocybe indica | Krishnamoorthy AS 2003 |
| 5      | Saw dust + rice husk | P.ostreatus | Singh MP et al., 2012 |
| 6      | Ramie stalks, Kenaf stalks | P.eryngii | Chinglung Li et al. 2016 |
| 7      | Rice straw, wheat straw, cotton straw, tea leaves, banana leaves | Pleurotus sp. | Ritika k. et al. 2017 |
| 8      | wheat straw | Agaricus bisporous | Ritika k. et al. 2017 |
| 9      | Rice bran, coffee pulp | Lentimula edodes | Ritika k. et al. 2017 |
| 10     | Tea leaves | Volvarella | Ritika k. et al. 2017 |
| 11     | saw dust | Ganoderma | Ritika k. et al. 2017 |
| 12     | composted sawdust + powdered Pineapple rind | P.ostreatus | Deborah L.N.M et al. 2017 |
| 13     | wheat straw + lathyrus straw | Calocybe indica | Anurag k. et al., 2018 |
| 14     | Pennisetum purpureum | Auricularia polytricha | Chih-Hung Liang et al. 2016 |
| 15     | Perilla stalks | P.ostreatus | Huizhen Li et al. 2017 |

All these lignocellulosic materials used as the substrate or supplement has shown to improve the yield, biological efficiencies and increase in the nutritional value of the various mushrooms.

8. Other Experimental Substrates

8.1 Used coffee grounds

There have been several efforts to reuse the waste products from production and consumption of coffee for the cultivation of mushrooms in different countries [38],[39]. It has been shown that using spent coffee grounds for mushroom cultivation has proved as an additional source of income for the coffee farmers of different countries [40],[41]. P.ostreatus is able to degrade about 86.6 % of the
cafeine in the given substrate at particular concentration [42], making this process not only economically lucrative but also ecofriendly.

8.2 Used tea leaves

India holds prominent position among the top producers of the tea in the world. With great production comes the great drawback of unutilized waste that causes pollution when leaches in the ground. Tea leaves are also cellulosic material and hence readily can be used for the cultivation of mushrooms. Autoclaved tea leaves were used waste along with wheat straw in different proportions for P. spadious and P. flabellatus. 3:1 and 1:1 ratio of TLW:WS gave best yield for P. flabellatus whereas 1:3 ratio gave best yield in P. spadious [43]. Tea leaves when used in the range of 40-60% along with cottonseed hulls gave best yield for P. oestrus [44]. P. sajor-caju gave better yield and fruiting body when sugarcane bagasse was added with tea leaves in ratio of 75:25 % and supplemented with 5% lime [45].

8.3 Banana leaves

Banana cultivation is done throughout the country and a lot of agro waste is generated after each harvest. As it is rich in lignocellulosic content it can prove to be good substrate for the cultivation of Pleurotus mushrooms. V. volvacea grew well in the banana leaf substrate at the same time fusig treated substrate showed high cellulose content making it a viable as ruminant feed [46]. Among three substrates tested banana leaves showed highest production and biological efficiencies for two strains of V. volvacea V99 and VVO [47]. The leaf and pseudo-stem of banana showed promising results for the cultivation of P. ostreatus [48].

9. Discussion

India with about 70% of rural households engaged in agriculture has about 275 million tonnes of total food grain production which also contributes to ample amount of lignocellulosic waste production. This lignocellulosic waste is utilized in little amount to not being utilized at all. India has a temperate climatic condition, which makes it favorable for the cultivation of mushrooms. Several rot fungi such as Pleurotus are efficient in growing in lignocellulosic waste and producing protein rich mushrooms. These lignocellulosic wastes can be used along with the substrates for the cultivation of mushrooms as, this will not only produce protein rich food to fight malnutrition but also at the same time help us in biodegradation of lignocellulosic waste. Several substrates such as wheat straw, rice straw, lathyrus straw, spent coffee grounds, spent tea leaves, banana leaves, vegetables, soybean straw etc. have shown to be promising in the cultivation of mushrooms. In developing countries like India, malnutrition is a prevalent issue. This issue can only be addressed by adequate diet and protein rich supplements and mushroom along with lignocellulosic wastes as substrate can play a major role in it.

10. Conclusion

Agro wastes containing of lignocellulosic materials can prove as a great substrate and supplements for the production of mushrooms as these are cost effective and readily available. Cultivation of mushrooms on agro wastes and spreading awareness about them will help the rural farmers in uplifting their socio-economic conditions.

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