

Mushrooms in the Bio-Remediation of Wastes from Soil

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Summary: Mushrooms have played a great role in the field of bioremediation. Mushrooms are saprophyte highly specialized group of macro-fungi with a distinctive fruiting body, and have a unique capacity for degradation of certain types of organic pollutants like lignocellulotic wastes and bio-sorption of heavy metals. The degradation of lignocellulotic wastes are initiated by the release of extracellular enzymes to the environment. The lignocellulolytic enzyme starts to degrade and breakdown the complex lignocellulotic wastes in to smaller and readily available molecules for their utilization. The present reviewed paper describes briefly the concerns regarding the extracellular mushroom enzymes, with having many potential applications in bioremediation of agricultural wastes, heavy metals and toxic organic compounds. Therefore, research is needed to develop understanding of integrated mushroom cultivation that optimizes mushroom utilization in the field of environmental remediation, while supporting other ecosystem services.

Keywords: Edible Mushroom, Bioremediation and Bio-degradation, Heavy metals

DOI: 10.7176/ALST/76-04

Publication date: September 30th 2019

1. Introduction

There is a growing concern about effective biological waste management system worldwide. Wastes are produced annually by different activities like agriculture, agro-industries, industries and municipal solid in the original form, without being processed. Annually tremendous amount of wastes and organic pollutants has been generated and are being released into the environment (water and soil) (Wijnhoven et al., 2007). Wastes are found in different forms; solid, liquid and semi-solid are the three major classes of waste. These wastes are being dumped or burnt without proper recycling, thus contributes to human health hazard, environmental pollution and finally global warming. Soils contaminated with heavy metals and/or organic pollutants are generally left abandoned for several years because they may not be safe for human health as well as agricultural production. Even though, a number of approaches have been used for the removal of heavy metals; none of these methods are as effective when compared with biological methods (such as the use of bacteria, fungi, algae, and plants) because they are expensive, fail to completely remove heavy metals and still more expensive.

The elimination of wide ranges of pollutants and wastes from the environment is therefore an absolute requirement to promote a sustainable development of our society with low environmental impact. Due to the magnitude of this problem and the lack of a reasonable solution, a rapid cost-effective ecologically responsible method of clean-up is greatly needed (Hamman, 2004). Various organisms like bacteria, fungi, algae, and plants have been used for decomposition of pollutants and cleaning up our environment (Leung, 2004). However, fungi are the most outstanding biological decomposer and; then i.e., they play a crucial role in converting these wastes into valuable products. They exist in a variety of habitats due to their versatile physiological nature thus found in acidic pH, temperature, oxygen concentrations, salinity and heavy metal concentrations (Chang and Miles, 1992). Mushrooms have been shown to degrade diverse type of wastes; organic and inorganic pollutants due to extracellular enzymes they possess thereby converting the waste and pollutants into food of high quality, flavor and nutritive value. Therefore, mushroom may be a promising fungi for environmental bioremediation.

Mushrooms belong to the family of Basidiomycetes commonly known as saprophytic fungi. Fruiting body of mushrooms consists of a stem (Stipe) with and a bearing cap (Pilues) a spore forming part (sporophore). Mushroom uptake nutrients from the substrate/soil via specious mycelium. The age and size of fruiting body determines the uptake of nutrients from the substrate or the soil (Das, 2005). The life span of fruiting body is only 10-14 days; "hence time taken consumed for uptake of these nutrients from the substrate is limited (Sharma *et al.* 2010) had suggested in their studies on nutrient contents in mushrooms that the uptake rate of nutrients can be correlated with the contact time. Nutrient concentrations in the fruiting bodies are affected by age of mycelium and interval between fructification (formation of fruiting (Sharma *et al.* 2010).

Mushrooms, are macro fungi with a distinctive fruiting body, are unique biota which assembles their food by secreting degrading enzymes and decomposing the complex organic materials on which they grow (the substrate) to generate simpler compounds for their nutrition (Chang and Miles, 1992). These substrate materials are usually by-products from industry, households and agriculture which are usually considered as wastes. These wastes, if carelessly disposed off in the surrounding environment by dumping or burning, will lead to series environmental pollution and consequently causing health hazards. However, they are actually resources in the wrong place at a particular time and mushroom cultivation can harness this waste/resource for its own beneficial advantage.

Mushroom also, can produce notable nutraceutical products, which have many health benefits. They provide people with an additional vegetable of high quality and enrich the diet with high quality proteins, minerals and vitamins which can be of direct benefit to the human health and fitness. The extractable bio-active compounds from medicinal mushrooms would enhance human's immune systems and improve their quality of life. Edible mushroom are highly nutritious and can be compared with eggs, milk and meat in terms of protein content. The composition of essential amino acids in mushroom is high and close to the need of the human body. Mushroom is easily digestible and it has no cholesterol content (Oei, 2003).

Mushroom is very environmental friendly, capable of converting the lignocellulosic waste materials into food, feed and fertilizers. It can be cultivated in containers like jars, basins, trays, plastic bags and other similar substances by providing artificial controlled conditions (Quimio, 1998). They are relatively fast growing organisms, can be harvested in 3 to 4 weeks after spawning. Thus, mushroom cultivation is short return significant biological decomposer and it plays a crucial role in converting wastes into valuable products. The spent substrate left after harvesting the mushrooms, with innumerable mushroom threads (collectively referred to as mycelia), which have been biochemically modified by the mushroom enzymes into a simpler and more readily digestible form, can be used as animal feed (more palatable), bio-fertilizer for soil fertility enrichment and biogas.

It has been revealed recently that mushroom mycelia can play a significant role in the restoration of damaged environments (myco-restoration) through myco-filtration (using mycelia to filter water or mycelia are used as a filter to remove toxic materials and microorganisms from water in the soil), myco-forestry (using mycelia to restore forests), myco-remediation (using mycelia to eliminate toxic waste, and myco-pesticides (using mycelia to control insect pests). These methods represent the potential to create clean ecosystem, where no damage will be left after fungal implementation (Stamets, 2005).

Besides their ability to degrade and convert lignocellulosic materials into human food, they can also act as effective bio-sorbent of toxic metals (Costa and Leite, 1991). Compared to green plants, mushrooms can build up large concentrations of some heavy metals, such as lead, cadmium and mercury in them as reported by Gast et al. (1988) in his studies on interaction of heavy metals with soil and mushrooms. This would suggest that fungi possess a very effective mechanism that enables them to take up some trace elements from the substrate (Latiff et al., 1996).

Soils contaminated with heavy metals and/or organic pollutants are generally left abandoned for several years because they may not be safe for human health as well as agricultural production. Moreover, to excavate and remove contaminated soil is relatively costly and environmentally unsafe procedure, if use chemical methods. But, mushroom cultivation is cost effective and friendly to environment, a biological process of converting wastes into valuable products. It is therefore, hoped that the advocating for mushroom farming will become a very important activity, which may lead to cleaning contaminated land.

Some of the comprehensive reviewed articles provide basic statues and different perspectives of mushroom cultivation and production viz. research history (Vetayasuporn, 2006) oyster mushroom cultivation on different cellulosic substrates, (Baysal and Peker, 2001) waste paper recycling, (Ashrafuzzaman, 2009) growth and yield of shiitake mushroom on sawdust from different plants, (Imtiaj and Ajjur, 2008) economic viability of mushrooms cultivation Rice straw, wheat straw, sugarcane waste, banana leaves, grass and sawdust. Moreover, it has been reported that mushroom is a bio-sorption of metals; it can absorb and accumulate heavy metals throughout their bodies. More than 90% of studies have been done on the production and cultivation of mushroom from agricultural wastes; few studies have been carried out on Khat (*Catha edulis*), water hyacinth (*Eicchorniacrassipes*Solms), and industrial sewage, domestic and municipal wastes for the production of mushroom. However, none of them pay attention to integrated mushroom production and cultivation for bioremediation of those wastes especially heavy metals from soil and water. Moreover, these wastes have been discarded along the roads and cause unpleasant odours and unhygienic conditions. So, there is an urgent need to retabulate the real involvement and benefits of this unique fungus in bioremediation/biodegradation of khat, industrial, sewage, domestic and municipal wastes and the mechanism of how to reduce heavy metals.

The presences of extracellular enzymes indicate diverse potential applications of mushroom in decomposition of wastes. Advancement in our knowledge about mushroom biodegradation may facilitate their wide applications for safe and sustainable environmental development and agricultural productivity. This article is aimed to highlight the biodegradation potential of mushroom and provides a moderate review on the progress made in mushroom research related to biodegradation and identifying the critical research needs for developing and implementing successful mushroom bioremediation as a model worldwide. The possible ways to maximize its multiple uses for mitigating the various pollutants were also suggested.

For instance the main reason that white-rot fungi are active to such a wide range of compounds is their release of extracellular lignin-modifying enzymes, with a low substrate specificity, so they can act upon various molecules that are broadly similar to lignin (Adenipekun and Lawal, 2012). The enzymes present in the system employed for degrading lignin include lignin-peroxidase (LiP), manganese peroxidase (MnP), various H₂O₂ producing enzymes (Kirk and Farrell, 1987) and laccase, although the three types of enzymatic activity are not

present in all lignolytic fungi.

Bioremediation using Mushroom

Bioremediation is one of the most promising alternatives for the control of environmental pollutions caused by heavy metals which have serious impact on human health and the environment. It is a process of degradation/removal of toxic organic materials, e.g. from oil spills, pesticides, and industrial waste, at the molecular level, converting them to more innocuous compounds. As stated earlier in this article, different approaches have been followed for the removal of heavy metals such as chemical precipitation, ion exchange, evaporation and extrapolating. However, none of these are effective compared to biological treatment since they are expensive and incomplete removal of metals. The ultimate goal of bioremediation is the full mineralization of contaminants, i.e. their transformation to CO₂, H₂O, N₂, HCl, etc. Heavy metal and radioactive cations, of course, cannot be decomposed but have their solubility lowered or reduced, e.g. by a change in oxidation state and (Singh et al., 2013), so that they become less harmfully in the ground, or might be removed by phytoremediation or myco-remediation, which involves harvesting the fungus.

Mushrooms have considerable potential in biotechnology and in bioremediation of waste materials. Gast et al. (1988) reported that mushroom have the potential to convert agricultural wastes and accumulated heavy metals namely Cd, Cu, Pb, Cr and Mn valuable products. The uptake rate varied from species to species (Akjn et al., 2010; Demirbas (2002); Zhang dan et al., (2008) (Table 1). For instance, *Agaricus bisporus* absorbs quit large amount of copper as compared to other metals; *pleurotus ostreatus* shows maximum uptakes of cadmium, minimum of mercury and zinc however, no uptake of lead; while lead accumulation by *Lepiotarhacodes* was very high (Das, 2005). Mushrooms also can degrade persistent xenobiotic compounds, e.g. *Pleurotus ostreatus*, *Trametes versicolor*, *Bjerkandera adusta*, *Lentinula edodes*, *Irpex lacteus*, *Agaricus bisporus*, *Pleurotus tuberregium*, *Pleurotus pulmonarius*. This would suggest that fungi possess a very effective mechanism that enables them to take up some trace elements from the substrate (Latiff et al., 1996).

According to Damodaran et al. (2011) the uptake rate of nutrients from the waste by mushroom is affected by the nature of substrates, pH, and species of mushrooms and to a certain extent the genus, biomass, and the presence or absence of heavy metals. The remediation process also varies accordingly the species; some species degrade the waste easily and quickly while others can break down the waste very slowly. The rate of remediation becomes maximal when there is a good supply of nutrients in the soil, e.g. N, P, K and other essential inorganic elements (Rhodes, 2013). *Pleurotus ostreatus* is able to degraded Benzo[a]pyrene but the presence of heavy metal cations and mediators such as vanillin and 2,2'-azinobis-(3- ethylbenzothiazoline-6-sulfonate) highly influence the degradation process. Bhattacharya et al., (2014), reported that 15 mM concentration of copper was found to best enhance the degradation (74.2%), progressively worsened as the Cu concentration increased. At the same time the extent of degradation was increased to 83.6 % when 5 mM of vanillin was included in the medium.

Costa and Leite, (1991) also reported the growth of mushroom spore in heavy metal contaminated soil. They were observed that natural biotechnology (Myco-remediation) for spinach plant cultivation under Pb metal stress condition: It was established that growth of fungal spore into mushrooms was compassionate or remove poisonous metals from the soil. It has been established the natural growth of the wild mushrooms in Pb contaminated environment to supports the growth of edible plants via directing or extracting contaminate through myco-remediation. This competently folds contaminants and absorb heavy metals by leading them to the fruit bodies of mushrooms. The content of heavy metals in sporocarp of edible mushrooms is ascribed in Table 1.

Table 1: Heavy Metal Content in Sporocarp of Various Tolerant Mushrooms

Mushroom Species	Metal content (Accumulated metals in sporocarp, mg/kg of dry weigh	References
<i>Agaricus bisporous</i>	Cu (107), Pb (21), Zn (57.2) Pb (2.4), Cd (3.48), Hg (0.6), Cu (5.22) Hg (0.03), Pb (0.28), Cd (0.78) Fe (31.3)	Isildak et al., (2003) Demirdas (2001)
<i>Boletus edulis</i>	Pb (0.96), Cd (1.03), Hg (0.13), Fe (31.1) Cu (4.7), Mn (2.9), Zn (26.2). Hg (32.4), Cu (66.4), Cd (6.58), Pb (3.03)	Tuzen et al. (1998) Kalac et al. (1996)
<i>Lepiota rhacodes</i>	Hg (8), Pb (66), Cd (3.7)	Kalac et al. (1996)
<i>Paxillus involutus</i>	Pb (1.6.0), Cu (57.0)	Kalac et al. (1991)
<i>Pleurotus sp.</i>	Pb (3.24), Cd (1.18), Hg (0.42), Cu (13.6), Mn (6.27), Zn (29.8), Fe (86.1). Pb (0.11), Cd (0.55), Hg (0.31), Fe (48.6),Cu (5.0), Mn (10.3), Zn (19.3)	Damodaran et al., (2011)
<i>Tricholoma terreum</i>	Cu (25), Zn (179), Mn (19), Fe (744), Co (2.6) Cd (0.56), Ni (5.6), Pb (4.4). Pb (2.4), Cd (1.6), Hg (0.06), Cu (35.8), Mn (24.8), Zn (48.0), Fe (169.0).	Dermirbas et al. (2001)
<i>Volverilia volvacea</i>	Hg &Pb (5-5.23), Cu 500	Damodaran et al., (2011)
<i>Volvariella murinella</i>	Pb (2.4), Cd (1.6), Hg (0.06), Cu (35.8)	Damodaran et al., (2011)
<i>Havlvela leucomelaena</i>	Pb (3.1), Cd (1.1), Hg (0.26), Cu (13.6); Pb (4.8), Cd (2.0), Hg (0.21), Fe (54.5)	Tuzen et al., (2003) Damodaran et al., (2011)
<i>Paxillus rubicondulus</i>	Pb (0.69), Cd (0.78), Hg (0.21), Fe (37.0), Cu (51.0), Mn (10.8), Zn (16.8)	Damodaran et al., (2011)

Source: Das (2005).

Genetic engineering in mushroom for enhanced bioremediation

Genetic engineering

So far there are no transgenic mushroom strains available commercially but several research groups are working towards that direction with good progress. Sequencing the mushroom genome is crucial since mushrooms are now regarded as being very important to the environment. This is in view of the fact that mushrooms help in degradation of agricultural wastes into less harmful substances, removal of heavy metals from waste flows and also play a role in production of biofuels (Thwaites *et al.*, 2007). Genetic engineering in mushrooms may provide opportunities to exploit for maximum benefit in the field of remediation technologies and to manipulate the tolerance, degradation potential of mushrooms against various organic and inorganic pollutants through introduction of desired genes. Thus, the development and application of genetic engineering of the native mushrooms will definitely offer more efficient and enhanced bioremediation of the pollutants viz. heavy metals, organics or co-contaminants, making the bioremediation more viable for environment remediation. Some points have to be put in mind such as biosafety assessment, risk mitigation, and factors of genetic pollution before using the genetically engineered fungi at field level including mushroom.

Other research needs related to mushrooms

It is well known that mushrooms are saprophytes and globally important in oxidizing the potent wastes. There is an urgent need to optimize the effect of different wastes for mushroom cultivation and production. The population dynamic and diversity of the mushrooms need to be studied with respect to edaphic and climatic conditions of environment. Moreover, integrated mushroom production and cultivation for bioremediation of wastes especially heavy metals from soil and water for sustainable agricultural production should be studied properly. Some of the commercial edible mushrooms and their origin are presented in table 2.

Table 2: Some common commercial mushrooms

Common name	Latin name	Origin
Button mushroom	<i>Agaricus bisporus</i>	Europe
Shiitake mushroom	<i>Lentinus edodes</i>	China
Oyster mushroom	<i>Pleurotus ostreatus</i>	Florida
Velvet stem mushroom	<i>Flammulina velutipes</i>	Japan
Paddy straw mushroom	<i>Volvariella volvacea</i>	China, Japan
Ear fungus	<i>Auricularia auricular</i>	China, Japan
Reishi mushroom	<i>Ganoderma lucidum</i>	Japan
Nameko mushroom	<i>Pholiotanameko</i>	China, Japan
white jelly mushroom	<i>Tremella fuciformis</i>	Taiwan
Truffle	<i>Tuber aestivum</i>	Europe, New-Zeland, Australia

Source: Chakravarty, (2011)

Common cultivated mushrooms

Although there are well over 300 genera of mushrooms and related fleshy basidiomycetes, only a few species of these fungi are cultivated commercially. This may be due to the fact that many of them are mycorrhizal and may not sporulate in the absence of the host. But many saprophytic species have been amenable to cultivation. Some of the more common cultivated species listed here (Table 2). The most common cultivated mushroom were oyster mushroom (*Pleurotus ostreatus*), button mushroom (*Agaricus bisporus*), Shiitake mushroom (*Lentinus edodes*), velvet stem mushroom (*Flammulina velutipes*), paddy straw mushroom (*Volvariella volvacea*), ear fungus (*Auricularia auricular*). However, other cultivated mushrooms serve as medicinal and flavoring agent are the Reishi mushroom (*Ganoderma lucidum*) which is used as an alternative medicine and also as flavouring agent in Japan; the Nameko (*Pholiota nameko*) grown in the orient and *Tremelia fuciformis* or white jelly fungi that is grown for use as food supplements in Taiwan. Varieties of *A. bisporus* that are grown commercially include the crimini and portabello. Truffles (*Tuber* species) live in close mycorrhizal association with roots of specific trees. They are considered a food delicacy and rated as one of the most expensive natural food in the world (Trappe *et al.*, 2007). Some of the edible mushrooms involved in the decomposition of various agricultural wastes are as presented in Table 3.

Table 3: Edible mushrooms involved in the decomposition of various agricultural wastes

Mushroom species	Growth substrates	Potential substrates	Yield
<i>Pleurotus flabellatus</i>	Mango, ack fruit, Coconut, Jam, Kadom, Mahogany, Shiris sawdust	Mango sawdust	150 gm
<i>Pleurotus ostreatus</i>	<i>Ficus carica</i> , <i>Albizia saman</i> , <i>Swietenia amahagoni</i> , <i>Leucaena leucocephala</i> , <i>Eucalyptus globulus</i> and mixture of all five tree sawdust	<i>Albizia saman</i>	373.4gm
<i>Volvariella volvacea</i>	banana leaves	banana leaves	2.5 kg
<i>Pleurotus ostreatus</i>	wheat straw (<i>Triticum aestivum</i>), maize stover (<i>Zea mays</i> L), thatch grass (<i>Hyparrhenia filipendula</i>) and oil/protein rich supplements (maize bran, cottonseed hull [<i>Gossypium hirsutum</i>])	Wheat straw	71gm

Source: Source: Chakravarty, (2011)

Conclusion

Mushrooms were discovered over a century ago; however, mushrooms have not been explored well. The research related to bioremediation potential of mushrooms is still in infancy stage. The production of new mushroom cultivars with novel and improved traits will provide the industry with options for solving food problems and increase the production efficiency. Improvement of tools available to the breeder, decoding mushroom genome and commercial pressure facing the industry can propel efforts for new strain development in the future. For better harnessing of mushroom in industrial application and bioremediation, a number of limitations need to be worked out such as integrated mushroom production and cultivation for bioremediation of wastes especially heavy metals from soil and water for sustainable agricultural production, metal uptake mechanism, antagonistic and synergetic effects of metals in uptake and the characters of these accumulates in molecular level. The metal uptake potential of mushrooms should be critically analyzed for health risk as

mushrooms have become a popular delicacy of modern world. With the combination of biotechnology and genetic engineering, mushrooms can be exploited for in situ bioremediation of a wide range of inorganic and organic pollutants.

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