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Review article

Mushrooms of the *Pleurotus* genus – properties and application

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Abstract: *Mushrooms of the Pleurotus genus are found naturally in forests in almost all latitudes where they are responsible for the decomposition of wood. These fungi are valuable to cultivate and eat, as they are source of valuable nutrients and healing ingredients. Mycelium of white rot is known for its bioremediation abilities, including the accumulation of heavy metals and chlorinated aromatic hydrocarbons. Mushrooms of the Pleurotus genus have also been found applicable in the biotransformation of unsaturated terpenoid compounds. These reactions involve hydroxylation at the allyl position and subsequent oxidation of the introduced hydroxyl group. The article presents a number of applications of various strains of fungi of the Pleurotus genus.*

Keywords: *Pleurotus, biotransformation, bioremediation, biodegradation.*

Introduction

Mushrooms of the *Pleurotus* genus, known as oyster mushrooms, are the main basic decomponenters of wood and plant residues. They grow mainly on decayed wood, but they are also capable of parasiting on living trees, infecting them with white rot [1]. Mycelium of white rot, i.e. oyster mushrooms, has bioremediation properties, cleansing contaminated soil with oil derivatives or polycyclic aromatic hydrocarbons or heavy metals [2].

Oyster mushrooms are found naturally in almost all latitudes except Antarctica, also in tropical and subtropical forests. Due to different growth conditions, mycologists have distinguished many species and types [3]. Mushrooms of this type are valuable to cultivate and eat. Their fruiting bodies are sources of easily absorbed proteins, carbohydrates, amino acids, B vitamins (thiamine, riboflavin and niacin), vitamin D and mineral salts (calcium, phosphorus, iron), characterized by low fat concentration [4-6]. In addition, they are sources of pro-health substances, including antibacterial, antifungal, immunomodulatory,

anti-inflammatory properties as well as reducing blood sugar and cholesterol levels [7].

Mushrooms of the *Pleurotus* genus fulfill a dual role in the environment. They are capable of colonizing and degrading a large number of lignocellulosic residues, and as edible and cultivated mushrooms they are a source of valuable nutrients and healing properties. These fungi require a shorter growth time compared to other edible fungi and are relatively rarely attacked by diseases and pests and can be cultivated [5,8].

Cultivation of oyster mushrooms

Mushroom *Pleurotus ostreatus* (Fr.) Kumm., used in cultivation for over 100 years, is now in the third place in the world in terms of production volume, after champignon mushrooms and *Lentinula edodes* (shiitake) [9]. The size and quality of yield depends on the growth conditions, i.e. the temperature and humidity of the substrate and air and acidity of the medium, as well as on the variety or species and the type of culture medium [10]. Oyster mushroom, as a saprophytic mushroom uses cellulose, hemicellulose and lignin for its growth, therefore is can be grown on wood, sawdust, straw of various species, maize settlements and other agricultural wastes. However, because it grows and yields faster on straw than on wood, its cultivation in Europe is most often carried out on straw, subjected to pasteurization [3, 11-13].

In China, where mushroom production is responsible for over 70% of the global market [9], the cultivation of *Pleurotus* spp. is mainly carried out on sawdust with various additions. Wastes from the production of cotton and cereal straw, especially rice are also used, but on a smaller scale [12, 14]. The availability of sawdust begins to be limited by the development of the Chinese poultry industry, therefore it is suggested to use wheat and rice straw, which is agricultural waste, currently unused. Additional enrichment of the straw of these species with the addition of cotton seed bags significantly improves the quality and yield of oyster mushroom compared to the surface of the straw itself [14].

Because China is also a potentate in the cultivation of herbs, research on the use of waste from their production for the cultivation of oyster mushroom are also conducted. The Jin team [15] used for this purpose, among others: sofora root (*Radix Sophorae flavescentis*), sarsaparilla rhizome (*Rhizoma Smilacis glabrae*) and other wastes coming from the pharmaceutical plant, added to the culture medium from corn sludges. They significantly increased the protein content and individual amino acids in fruiting bodies. They also increased the antioxidant activity of *P. ostreatus* fruiting bodies.

In Africa, oyster mushrooms are grown mainly on sawdust, with the addition of rice straw. Looking for nutritional supplements, Narh Mensah and his team [16] used the addition of powdered pineapple skins. The addition of 2 and 5% positively influenced the yield and nutritional value of the cultivars EM-1 cultivated this way. Solutions regarding the use of waste additives other than

those used so far contribute both to increased efficiency and productivity as well as to environmental protection, reducing the amount of waste deposited.

Mushroom cultivation is usually carried out under fully controlled conditions. For oyster mushroom during mycelial growth in the culture medium, the temperature should be 24ºC, relative humidity 85-95%, CO_2 content from 5000 to 20000 ppm. During molding the initial temperature of the air should be 10-15ºC, relative humidity 95-100%, CO_2 content below 1000 ppm, with light access in the range of 1000-2000 lux. When growing sporocarps, the air temperature should be in the range of 10-21ºC (depending on the species), and air humidity in the range of 85-90% [13]. Depending on the technological advancement of facilities used in mushroom cultivation, these conditions are met to a lesser or greater extent. However, greater precision of the crop increases its productivity as well as healthiness [8].

Both the culture medium and growth conditions play a huge role in the cultivation and yielding of oyster mushroom. They are also dependent on the degree of hygiene in the production process because the cultivation of oyster mushrooms, like in champignon mushrooms, is an intense monoculture. Failure to comply with hygienic standards results in increased occurrence of fungal and bacterial diseases and pests [3, 8, 13]. Although, compared to other cultivated mushroom species, oyster mushrooms are less frequently attacked by diseases and pests [8], however, the use of protection measures is an important element in their production.

The source of medicinal and nutritional substances

The fruiting bodies of fungi of the *Pleurotus* genus are a rich source of micro and macro elements, such as copper, iron, zinc and sodium, potassium, magnesium and phosphorus. Their amount depends to a large extent on the species, age and size of the fungus, as well as the growing conditions. The mushroom pilei contain primarily potassium and phosphorus, whose content varies between 10-20 mg/100 g. The content of other minerals is smaller, on the order of a few percent. Mushrooms of the *Pleurotus* genus are also characterized by a high content of valuable nutrients. These are protein (20-25%), carbohydrates (40-46%), amino acids (20-40%) and fiber (10-20%). Significantly, the fat content is very low, it is within 10-20% of the dry matter [17].

In Chile there are two species of oyster mushroom: *P. ostreatus* and *Pleurotus sutherlandii*, growing on the endemic *Nothofagus* tree. In one of them – *P. sutherlandii* – lovastatin **1** (Figure 1) has been identified, a compound that reduces cholesterol in the blood [4]. Successive researchers tested *P. ostreatus* using two variants of fungal growth. In the first case fruiting bodies were collected directly from the trees. In the second case, fruiting bodies of fungi that were grown on wheat straw were harvested under greenhouse conditions. After extracting with methanol and ethyl acetate, the content of lovastatin **1** in the tested fungi was determined. It turned out that the content of this compound is

0.4 to 2.07% on a dry weight in the case of cultivated fungi and 0.7 to 2.8% in the case of naturally growing fungi. This means that much better source of lovastatin **1** is oyster mushroom growing in the natural environment [18].

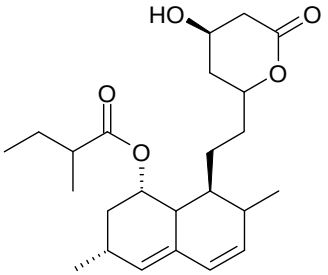

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Figure 1. Formula of lovastatin

Two strains *P. ostreatus* DSM 1833 and *Pleurotus sajor-caju* CCB 019 were tested as a part of the research on increasing the nutritional value of *Pleurotus* fungi. Rice and banana straw were used as a plant material for growth. As a result of the research, it was found that the type of culture medium did not affect the carbohydrate content in both strains. *P. ostreatus* fruiting bodies were characterized by lower humidity using rice straw than banana straw, whereas in case of *P. sajor-caju* fruiting bodies it was the opposite. The total fat content was higher in *P. ostreatus* than in *P. sajor-caju* when rice straw was used. Both strains had a higher total fiber content when grown in rice straw. In turn, the protein content was higher with the use of banana straw [5].

Biotransformation

Mushrooms of the *Pleurotus* genus have found usage in biotransformation of terpenes. Lyophilized strain of *Pleurotus sapidus* suspended in Tris-HCl buffer transformed (+)-valencene **2** into (+)-nootkaton **5**. Additional products of this reaction were two allyl alcohols α-nootkatol **3** and β-nootkatol **4**. Further experiments showed that only β-nootkatol **4** was an intermediate product in the oxidation of valencene **2** to nootkaton **5**. As a result of genetic tests, it was confirmed that the oxygenase protein present in *P. sapidus* is similar to the lipoxygenases found in *Aspergillus ochraceus*, *Aspergillus fumigatus*, *Gibberella moniliformis* and *Laccaria bicolor* [19]. Lipoxygenases belong to the group of dioxygenases containing in their structure an iron ion, without the presence of a heme structure. Such enzymes are found in plants, animals, bacteria and fungi. Lipoxygenases are responsible for the oxidation reactions of double bonds of polyunsaturated fatty acids. In this case, an alternative two-step mechanism of enzyme action was proposed, consisting of introducing the hydroxyl group in the allyl position and then oxidizing this group to the ketone.

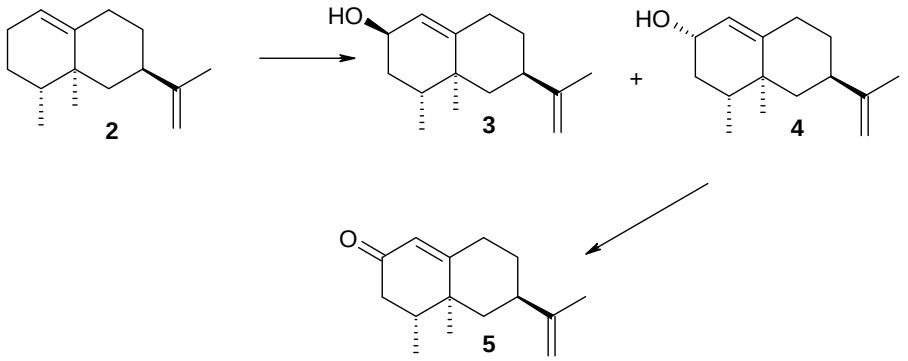


Figure 2. Biotransformation of valencene

Studies on biotransformation of valencene were continued by subsequent researchers. They found that the intermediate products of hydroxylation of compound **2** are hydroperoxides, the formation of which depends on the presence of valencene dioxygenase in *P. sapidus*. The mechanism proposed by the authors is similar to the catalytic mechanism of action of known lipoxygenases. Dioxygenases, such as lipoxygenases, initiate a reaction from detachment of the hydrogen atom from the substrate molecule. The resulting hydroperoxides are stabilized by introducing the hydroperoxide group in the allylic position. The reaction mechanism assumes the formation of two intermediate forms **A** and **B** (Figure 2) [20].

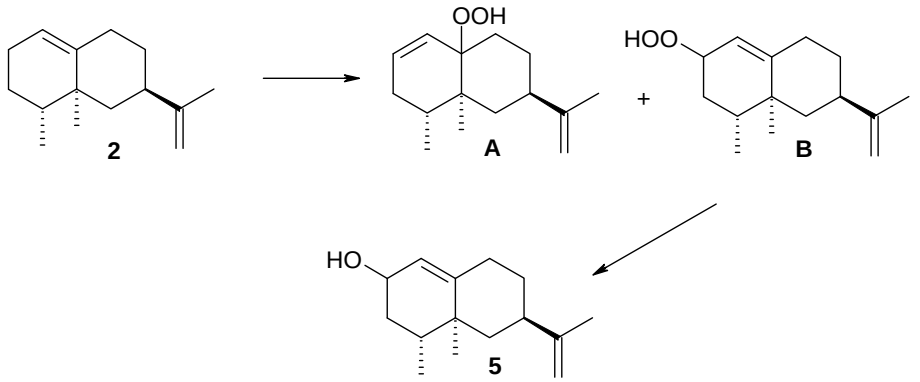


Figure 3. Biotransformation of valencene

The *P. sapidus* strain was also used to carry out biotransformation of kar-3-ene **6**. The transformations were carried out in the aqueous culture of the strain for 4 hours. After this time, compounds **7**, **8** and **9** were obtained as the main products (Figure 4). It was found that the mechanism of formation of these compounds is consistent with the action mechanism of dioxygenases presented

earlier. The formation of appropriate ketones is preceded by the formation of transitional hydroperoxide forms and allyl alcohols [21].

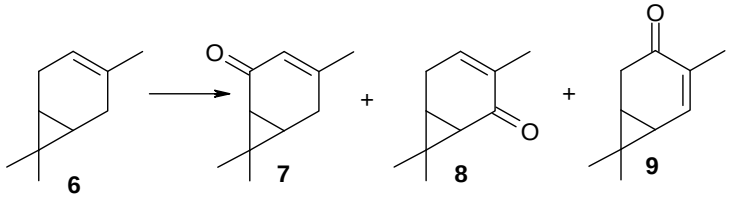


Figure 4. Biotransformation of car-3-ene

The next substrates used for biotransformation performed with the *P. sapidus* strain were allyl spiroethers **10**, **11**, **14** and **15**. The lyophilisate *P. sapidus* suspended in Tris buffer was used for the reaction. Biotransformation were carried out at room temperature for 48 hours. It was found that the structure of the substrate significantly influences the course of the reaction. Compounds **10** and **11** were transformed into the corresponding lactones **12** and **13**, while their structural analogies **14** and **15** did not undergo any transformations. The conversion rate for compounds **10** and **11** was 77% and 99%, respectively (Figure 5).

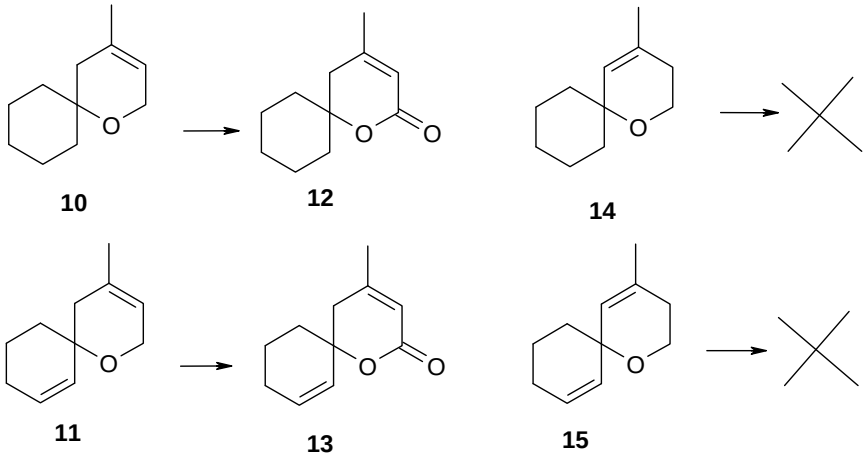


Figure 5. Biotransformation of spiroethers

Under similar conditions biotransformation of vitispirane **16** naturally occurring in grape juice was carried out. As a result, ketoalcohol **17** and two diastereoisomeric diols **19** and **20** were obtained. The 80% of the substrate was converted. The mechanism proposed by the authors assumes that the intermediate product of this reaction could be epoxide **18**, converted by hydrolysis to diols **19** and **20**. In turn these compounds could be oxidized to product **17** (Figure 6) [22].

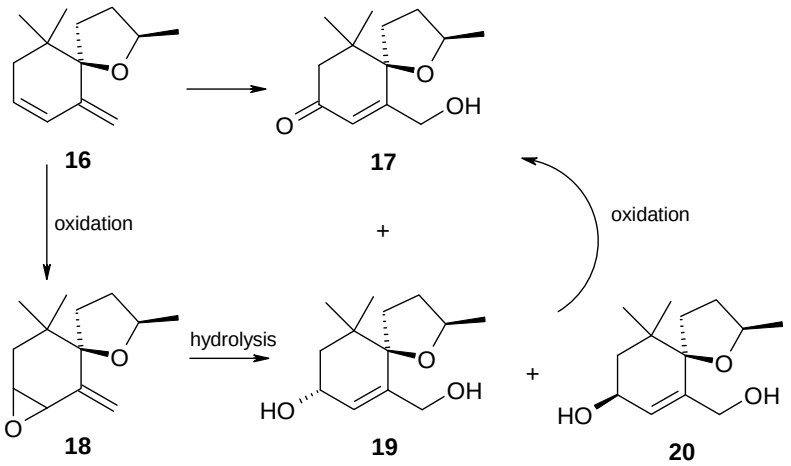


Figure 6. Biotransformation of vitispirane

Pleurotus euosmus DSM 5331, *P. ostreatus* DSM 5339, *P. sajor-caju* DSM 1020 and *P. sapidus* DSM 8266 strains were used to carry out biotransformation of *R*-(+)-limonene **21**. It was found that only *P. sapidus* was able to hydroxylate this compound to an equimolar *cis*/*trans* mixture of carveols **22** after 7 days of the process. The same strain (*P. sapidus*) was then used to oxidize enantiomeric carveols to carvone **23**. Studies have shown that *trans*-(-)-carveol was converted to enantiomerically pure *R*-(-)-carvone during 4 days with 94% efficiency. Under the same conditions, *cis*-(+)-carveol was oxidized to *S*-(+)-carvone only in 55% (Figure 7) [23].

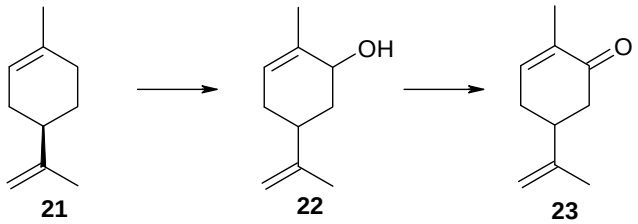


Figure 7. Biotransformation of *R*-(+)-limonene

Heptachlor **24** is a representative of synthetic insecticides, used in the 60's and 70's of the last century. This compound was used to exterminate termites and insects living in the soil. Heptachlor may oxidize to epoxide, which is more durable and more toxic than the parent compound. Both compounds are found in soil all over the world. Heptachlor has toxic effects on the human body by attacking the central nervous system. Because fungi of the *Pleurotus* genus are able to biodegrade polychlorinated compounds, this time it was decided to use them for biotransformation. Heptachlor **24** and its epoxide **25** were used as substrates, whereas *P. ostreatus* BM9073 as a bioreagent. This microorganism

was grown on several culture media. The medium composed of 1% glucose, 1.2 mM ammonium tartrate and 20 mM sodium acetate (HN) and medium containing potato dextrose broth (PDB) proved to be the best. In both cases, complete conversion of heptachlor was observed after 14 days of incubation. The biotransformation products were heptachlor epoxide **25** (about 65%), chlordene **26** (about 23%) and 1-hydroxy chlordene **27** (about 2%). The heptachlor epoxide **25** was in turn transformed into diol **28** at 8% and 31% respectively. The resulting final products as hydroxy derivatives may show less toxicity than the starting compounds (Figure 8) [24].

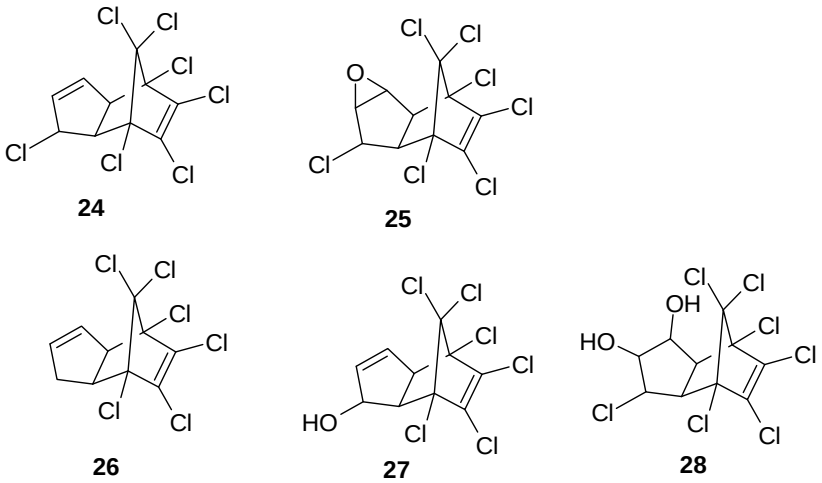


Figure 8. Heptachlor and its derivatives

Bioaccumulation of heavy metals

The characteristic feature of white rot fungi, to which *P. ostreatus* belongs, is the ability to accumulate heavy metals. *P. ostreatus* has been tested for its potential to completely remove copper, nickel, zinc and chromium from the water. A solution consisting of 23.56 mg of Cu (II), 54.83 mg of Ni (II), 42.87 mg of Zn (II) and 93.54 mg of Cr (VI) dissolved in 1 dm^3 of water was used in the experiment. Various parameters were examined, such as the effect of pH, the amount of biomass, equilibrium time, mixing intensity, temperature and initial concentrations of metal ions. It was found that the maximum adsorption of Ni (II), Cu (II) and Zn (II) ions occurred in the pH range 4.5-5.0, while for Cr (VI) ion the best results were obtained at a pH of 2.5. Studies on the effect of biomass amount showed that during increasing the biomass from 0.1 to 0.3 g, an increase in efficiency in removing metal ions was observed. Further increasing the amount of biomass from 0.4 to 0.8 g had no significant effect on biosorption. Observations conducted during the process over time proved that in the first 15 minutes there was a rapid increase in the sorption of metal ions due to their rapid uptake by fungi. Then the rate of metal removal increased gradually and

reached its maximum value within 120-150 min. Biosorption took place in two phases. The first phase consisted in the rapid storage of metal ions on the surface of the fungus, while the second involved the slow transfer of metal ions to the cytoplasm of the cell by membrane transport or slow intracellular diffusion. Temperature in the range of 20-45°C had no significant effect on biosorption. The maximum fungal biosorption capacity was 8.06, 20.4, 3.22 and 10.75 mg/g for Cu (II), Ni (II), Zn (II) and Cr (VI), respectively [25].

During the next tests, the ability of the immobilized *P. ostreatus* to remove Cd (II) ions from contaminated water was tested. The biosorption capacity and biosorption rate were determined depending on pH, temperature and cadmium ions concentration. The tests showed that both the capacity and rate of biosorption reached the value of 70.3% at pH 6. The increase of temperature from 5ºC to 30ºC resulted in increased capacity and biosorption rate from 7.3% to 72.6%. A different situation was observed when studying the effect of the initial concentration of cadmium ions. The experiment was carried out by increasing the initial concentration of cadmium from 25 to 200 mg/L. It has been found that the biosorption capacity increases rapidly up to a concentration of 150 mg/L, and then remains constant. In turn, the biosorption rate decreases from 97% (25 mg/L) to 40% (200 mg/L). The best parameters allowing removal of 87% of cadmium from the water tested were: initial concentration of Cd (II) ions of 200 mg/L, pH = 6 and temperature of 25°C [26].

In another experiment, soil originating from a municipal waste landfill was used. The studied soil, fungi *Pleurotus ostreatus* along with straw used as a source of nutrients were laid out with qualities. The whole was incubated for 22 days. It was found that after this time 68% of lead and 81.25% of nickel were removed from the soil [27].

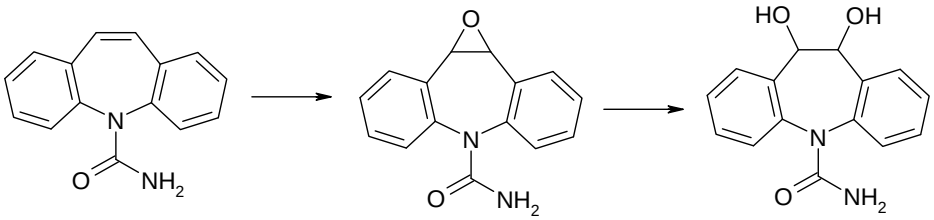
Cobalt is a micronutrient important for soil microorganisms and crop plants. However, when the level of cobalt exceeds the acceptable limit, it can lead to soil and crop contamination. One of the methods of removing excess cobalt from the soil is to use the so-called spent mushroom substrate. The tested Chinese soil samples (*Brassica chinensis L.*) were grown on the tested soil samples containing the spent fungal substrate and cobalt. Within 28 days, the phyto-availability of cobalt in the soil and its accumulation in cabbage were determined. It was found that the best results were obtained when the concentration of spent fungal substrate ranged from 8.86 to 9.51 g kg-1, the phyto-availability of cobalt in the soil reached a minimum, while the biomass of cabbage reached a maximum. This means that the used fungal substrate from *Pleurotus ostreatus* effectively reduces the cobalt availability – limiting the possibility of transferring this metal to the human body through the consumption of food [28].

Another method was the use of *P. ostreatus* immobilized on bentonite to remove traces of heavy metals from the water. In the experiment, 200 mg of dry mycelia mixed with 2 g of bentonite was used. The finished mixture was placed in a chromatographic column. The solutions of test metals containing 2.5-25 μg

of Cd (II) and Pb (II) in 100 ml of tap water of the selected pH were passed through the column and the degree of their biosorption was determined. The optimum pH value for both metals was pH = 5. The degree of sorption of metal ions ranged from 85% to 90%. It has also been found that the optimal solution flow rate through the column was 2.5 mL/min and the required contact time of the solution with the substrate was 30 minutes. Once filled, the column can be used 20 times without significant changes in the recovery of metal ions. The described method may be a cheaper alternative to activated carbon used to purify water from trace amounts of heavy metals [29].

Biodegradation

Carbamazepine (CBZ) **29** is an anticonvulsant used mainly for the treatment of epilepsy. This compound is slightly removed in municipal wastewater treatment plants and as a result it accumulates in the natural environment. Unfortunately, it has toxicological effects on aquatic organisms. The use for biodegradation of *Cunninghamella elegant*, *Umbelopsis ramanniana*, *Trametes versicolor* or *Ganoderma lucidum* proved that these fungi are able to metabolize carbamazepine in 25-60% during 17 days of incubation. In the next experiment strains of the *Pleurotus* genus: Florida N001, PC9 and Florida F6 were tested. It was observed that these strains degraded CBZ in the range from 48 to 99%. The best result was observed for the PC9 strain, so it was used for further research to determine the mechanism of CBZ biodegradation. Previous studies have suggested that two enzymatic mechanisms may participate in the oxidation of CBZ: the ligninolytic system of white rot fungi and the CYP450 monooxygenase system [30, 31]. *P. ostreatus* PC9 was grown in various media to check the accuracy of any of the above hypotheses. When both CYP450 (cytochrome P450) and MnP (manganese peroxidase) were active, 99% of the added CBZ was eliminated from the solution and converted to 10, 11-epoxycarbamazepine **30**. Inactivation of CYP450 or MnP also resulted in the removal of CBZ, but at slower pace. In the case of absence of both systems, only 30% CBZ was removed during the 32 days of incubation. This means that both systems participate in the oxidation of CBZ. The biotransformation product epoxide **30** exhibits pharmacological activity similar to the activity of the parent compound. However, after the activation of the CYP450 and MnP enzyme systems, it was observed that the epoxide formed in the first step is gradually converted to 10, 11 *trans*-diol **31**. Such a compound may be bioavailable to other microorganisms (Figure 9) [32].



29 **30** **31**

Figure 9. Biodegradation of carbamazepine

Aflatoxins are highly carcinogenic secondary metabolites that can contaminate about 25% of crops, particularly in Africa and Asia. White rot fungi are able to decompose aflatoxins *in situ* and *ex situ*. Therefore, studies were carried out to determine the ability of *P. ostreatus* to degrade aflatoxin B1 (AFB1) **32** (Figure 10) in naturally contaminated maize using standard cultivation techniques. It was found that the growth of fungi was not inhibited by AFB1 contaminating maize in an amount from 25 ng/g to 2500 ng/g. In addition, no detectable amount of aflatoxin was observed in 100 × concentrated extracts of *P. ostreatus* fungi grown on AFB1 contaminated corn, regardless of the *P. ostreatus* strain used or the initial level of AFB1. This means that microorganisms, in particular white rot fungi, can be used to degrade aflatoxin in crops intended for the consumption of livestock [33].

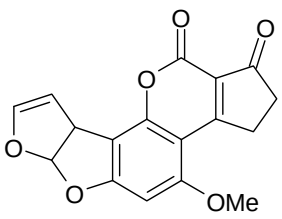


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Figure 10. Formula of aflatoxin B1

Bisphenol A (BPA) **33** is used for the production of plastics, mainly polyesters, including polycarbonates, epoxy resins, polyethers, polysulphones. This compound causes pollution of the natural environment, accumulating both in water and in soil. In its removal from the environment, *P. ostreatus* may be useful. The *P. ostreatus* O-48 strain was tested *in vivo* and *in vitro*. For the *in vivo* experiment, a homogenized mycelium suspended in 20 ml of medium containing glucose, peptone yeast extract and mineral salts to which 0.4 mM BPA was added was used. After 12 days of transformation in stationary culture, about 20% of BPA remained from the initial amount of compound. During the *in vitro* experiment, the enzyme MnP (manganese peroxidase) produced by *P. ostreatus* was used for the study. This enzyme was obtained from the liquid *P. ostreatus* O-48 culture. The results of the experiments proved that BPA **33** was

metabolized to phenol **34**, 4-isopropenylphenol phenol **35**, 4-isopropylphenol **36** and hexestrol **37** (Figure 11) [34].

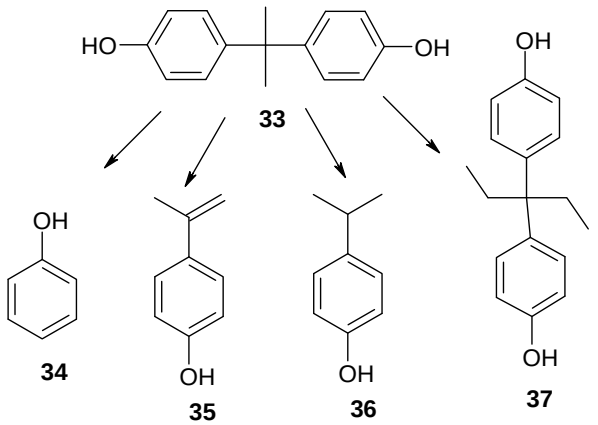

Figure 11. Biodegradation of bisphenol A

Fluoranthene **38** (Figure 12) is a representative of polycyclic aromatic hydrocarbons, compounds that pose a threat to the natural environment. Many of them are potentially genotoxic and carcinogenic. The main source of such compounds is the burning of fossil fuels and industrial processing, but also forest fires. It is known that white rot fungus is able to catalyze degradation of polycyclic aromatic hydrocarbons to quinones and hydroxylated aromatic compounds. Therefore, the *P. ostreatus* HP-1 strain was tested in a forest near Gujarat, India. It was found that this fungus is able to grow on a medium containing 50 mg/L of fluoranthene. The tests showed that after 54 days of transformation, 54.09% of fluoranthene was degraded. It was also found that this compound was broken down into an aliphatic compound [35].

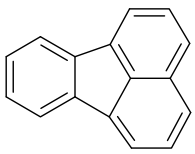

Figure 12. Formula of fluoranthene

Atrazine (2-chloro-4-ethylamino-6-isopropylamine-s-triazine) **39** is an herbicide frequently used to control weeds in sugar cane crops. Methods of degradation of atrazine through the *P. ostreatus* strain INCQS 40310 was used for the degradation of atrazine. Various types of culture media were used for growing this fungus. In the first approach, a standard PDA medium consisting of 2 g glucose, 1 g peptone and 2 g yeast extract in 1 dm^3 of water was used. In this case, the degree of degradation of atrazine was 39.0% after 15 days. Then, the

effects of changes in the amount of nutrient components and the use of various inorganic salts were tested. Preliminary studies have shown that the best effect was achieved by increasing the glucose concentration to 8 g/L and adding $ZnSO_4$ (0.002 g/L), $FeSO_4$ (0.001 g/L) and $MgSO_4$ (1 g/L) to the medium. The degree of degradation of atrazine after 15 days of biodegradation increased to 71%. Slightly worse result (63.3%) was obtained for medium with increased amount of peptone (5 g/L) with addition of $FeSO_4$ (0.001 g/L), $MgSO_4$ (1 g/L) and $CuSO_4$ (0.5 g/L). In the following experiment, the best medium was subjected to further tests, namely the effect of only two inorganic salts of $FeSO_4$ and $MnSO_4$ at different concentrations. It turned out that the best result was obtained when 0.001 g/L $FeSO_4$ and 0.05 g/L $MnSO_4$ were used. Under these conditions, after 94 days 94.5% atrazine was degraded. In addition, it has been found that as side products of the degradation of atrazine **39**, hydroxylated and chlorinated compounds were formed: 2-chloro-4-amino-6-isopropylamine-s-triazine **40**, 2-chloro-4-ethylamino-6-amino-s-triazine **41**, 2-chloro-4,6-amino-s-triazine **42**, 2-hydroxy-4-amino-6-isopropylamino-s-triazine **43**, 2-hydroxy-4-ethylamino-6-amino-s-triazine **44** (Figure 13) [36].

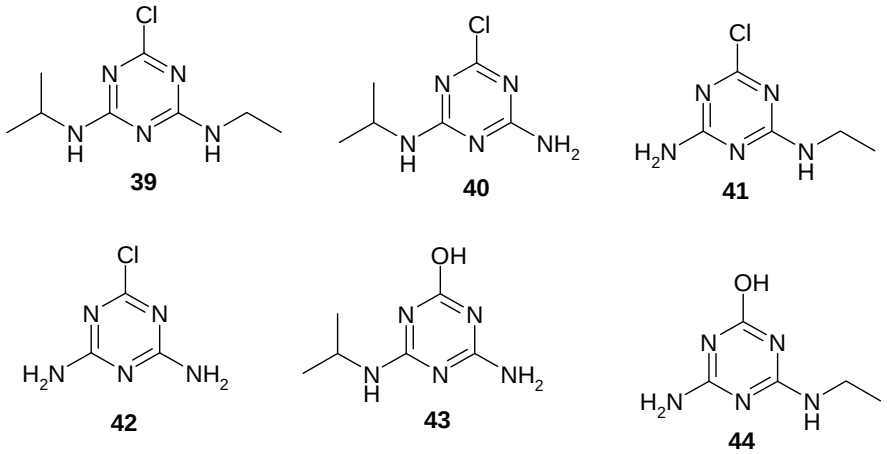

Figure 13. Atrazine and products of its degradation

DDT (1,1,1-trichloro-2,2-bis-(4-chlorophenyl)-ethane) **45** was the first of synthetic pesticides that was used on a large scale as a plant protection product from the 1940's to the 1970's of the last century. This compound has a long half-life of 2-15 years in soil. Its decomposition products are mainly DDE (1,1-dichloro 2,2-bis-(4-chlorophenyl)-ethane) **46** and DDD (1,1-dichloro-2,2-bis-(4-chlorophenyl)-ethylene) **47** with properties similar to DDT, but with even longer disintegration time. This compound is very harmful to higher organisms, because it damages the nervous system, destroys DNA in the blood cells and interferes with the synthesis and metabolism of endogenous hormones.

On a medium consisting of japanese cedar, sawdust, rice bran and water, mycelium of *P. ostreatus* was inoculated, and the whole was incubated at 20°C

for 28 days. After this time, so-called fungal substrate (SG), which was the substrate along with the growing mycelium and SMW, i.e. fresh waste remaining from the substrate after collecting the fungi were obtained. Both the fungal substrate and SMW were then used to study the utilization of DDT. It was found that 37% (using SG) and 48% (using SMW) of this pesticide were degraded during 28 days of incubation. In turn, the mineralization was 4.4% and 5.1% DDT within 56 days, respectively. Subsequent attempts concerned purification artificially contaminated with soil DDT. In this case, it was found that SMW degraded DDT in 40% and 80% in the sterilized and inexperienced soil, respectively. In such soil samples, during 56 days, the mineralization of DDT in the amount of 5.1% and 8.0% occurred. The tested pesticide was degraded to compounds **46**, **47** and additionally 2,2-bis-(4-chlorophenyl)-acetic acid **48**, 1-chloro-2,2-bis-(4-chlorophenyl)-ethylene **49**, 1-chloro-2,2-bis-(4-chlorophenyl)-ethane **50** (Figure 14) [37].

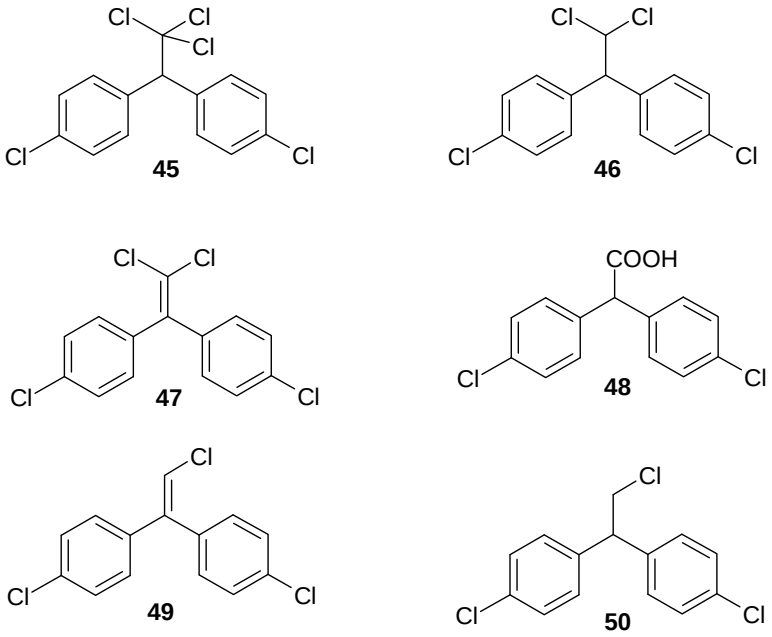

Figure 14. DDT and its derivatives

Aldrin **51** and its metabolite dieldrin **52** are compounds that permanently contaminate soil in many parts of the world. Considering the potential risks associated with these pollutants, an effective method of their degradation is needed. The *P. ostreatus* strain was used for biodegradation. As the culture medium the medium with low nitrogen content (1% glucose, 20 mM sodium acetate and 1.2 mM ammonium tartrate), medium with high nitrogen content (1% glucose, 20 mM sodium acetate and 12 mM ammonium tartrate) and broth with dextrose potato were used. Depending on the type of medium used,

P. ostreatus eliminated 25%, 72% and 100% of aldrin **51**, respectively, during the 14-day incubation period. The main metabolite was dieldrin **52**, besides 9-hydroxydieldrin **53** and 9-hydroxyaldrin **54** were present in the medium. The proposed biotransformation pathway for aldrin **51** was the epoxidation of a double bond followed by the hydroxylation of the epoxide ring. *P. ostreatus* was also capable of dieldrin **51** degradation. In an environment with a low nitrogen content, high nitrogen content and in dextrose broth, during the 14-day incubation period, about 3, 9 and 18% of dieldrin were eliminated, respectively. In the third variant (dextrose broth), 9-dihydroxydieldrin **53** was detected as a metabolite. The formation of hydroxylated derivatives is the first step to obtain less toxic metabolites (Figure 15) [38].

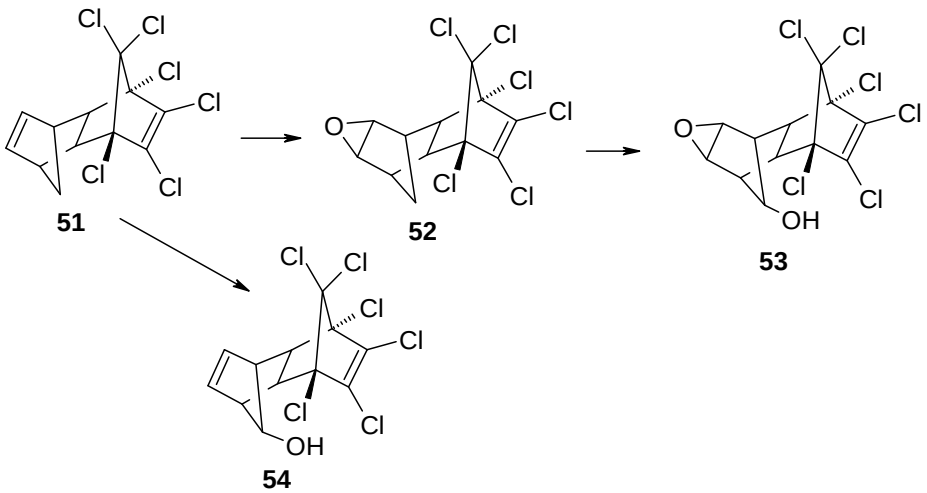

Figure 15. Biodegradation of aldrin

Phthalates are a group of persistent chemicals used primarily as additives to plastics in order to increase their elasticity. Phthalates are easily released from plastics into the environment by direct release, migration, leaching and abrasion as they are not chemically related. Long-chain phthalates, such as di-(2-ethylhexyl)-phthalate (DEHP) **55**, are used in polyvinyl chloride polymers. DEHP and its metabolite mono-(2-ethylhexyl)-phthalate **56** have toxic effects on the liver, reproductive system, heart, kidneys and lungs in both primate and rodents (Figure 16).

The *P. ostreatus* strain was used for the biodegradation of DEHP. Three types of medium were used. The control medium consisted of glucose, yeast extract, mineral salts (KH_2PO_4, $MgSO_4 \cdot 7H_2O$, K_2HPO_4, $CuSO_4 \cdot 5H_2O$, $FeSO_4 \cdot 7H_2O$, $MnSO_4$, $ZnSO_4 \cdot 7H_2O$) and Tween 80. Two other medium contained in addition to the above ingredients DEHP in 500 mg/L and 1000 mg/L. After adding the growing mycelium *P. ostreatus* to the medium, the whole was incubated for 21 days. The highest biomass production was observed in the medium enriched

with 1000 mg DEHP, and slightly lower in the medium containing 500 mg DEHP. It follows that *P. ostreatus* uses high concentrations of DEHP as a source of carbon and energy. Phthalate was degraded completely after 504 hours. DEHP can be metabolized through three pathways; the de-esterification pathway, the oxidation pathway and the oxidation-hydrolysis pathway, respectively forming phthalic acid, acetic acid and butanediol [39].

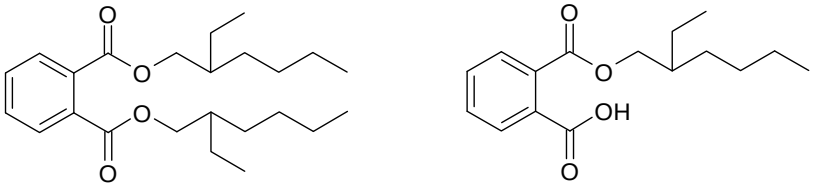

Figure 16. Phthalates

Summary

Mushrooms of the *Pleurotus* genus can be found in many applications in the modern world. They are used by the food industry as valuable edible mushrooms. The pharmaceutical industry can use them as a source of medicinal substances. Because of them, new oxygen derivatives of terpenoid compounds can be obtained. What is equally important, these fungi contribute to the purification of the natural environment from harmful compounds produced by man.

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