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Evaluation of cadmium accumulation in pink oyster mushrooms cultivated on the cadmium contaminated substrates and health risk analysis

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Abstract: Pink oyster mushrooms are rich in protein, dietary fibre, vitamins and minerals, making them a great addition to any diet. However, pink oyster mushrooms have the ability to accumulate high concentrations of heavy metals, some of which, such as cadmium, can cause adverse effects on human health. The aim of this study was to evaluate the ability of pink oyster mushrooms to absorb Cd from substrates contaminated with Cd and to assess the human health risks associated with the consumption of these mushrooms. An experiment was carried out in a completely randomised design and included four treatments (four Cd contamination levels i.e. 0, 20, 50 and 100 mg·kg^{-1}) with three replications. Cd accumulation in mushrooms increased with increasing Cd content in substrates and ranged from 1.8 mg·kg^{-1} (non-contaminated substrate) to 23.8 mg·kg^{-1} of dry mass (substrate contaminated with 100 mg·kg^{-1} of Cd). On the other hand, total mushroom yield showed a decreasing trend with increasing Cd levels in substrates. The results of the present study suggest that pink oyster mushrooms possess the capability to absorb Cd from the substrate in which they grow. The obtained results for target hazard quotient (*THQ*) of Cd point to the conclusion that the consumption of mushrooms cultivated on the Cd-contaminated substrates could produce negative health effects.

Keywords: consumption; edible mushrooms; heavy metals; toxicity

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Pink oyster mushrooms (*Pleurotus djamor* (Rumph. ex Fr.) Boedijn) are characterised by deepest pink colour, unique shape, delicious flavour, short production cycle and high yield. In addition, they are rich in protein, dietary fibre, non-starchy carbohydrates, vitamins and minerals, making them a great addition to any diet. They also contain numerous bioactive compounds with high health-promoting effects, leading to their increasing cultivation worldwide (Raman et al. 2020; Chowdhury et al. 2024). However, pink oyster mushrooms, like other mushrooms, have the ability to accumulate elevated concentrations of heavy metals (Mleczek et al. 2021), some of which, such as cadmium, chromium, mercury and lead can cause adverse effects on human health even at low levels (Dowlati et al. 2021).

The accumulation of heavy metals in mushroom fruiting bodies is primarily affected by the heavy metal concentrations in the mushroom growing substrate, suggesting that mushrooms do not have effective mechanisms to block or avoid the uptake of heavy metal ions from the growing substrate in which they grow (Ab Rhaman et al. 2021). From this point of view, it is realistic to expect that heavy metal concentrations in mushrooms increase sharply with increasing heavy metal concentrations in mushroom growing substrate, and this hypothesis is being confirmed by a number of case studies (Demirbaş 2002; Kapahi and Sachdeva 2017; Gupta et al. 2021). Conversely, several studies have indicated that mushrooms can accumulate varying concentrations of heavy metals even when cultivated on the same substrate. This suggests that the uptake of metals is influenced not only by the concentration of heavy metals in the growth medium and environmental factors but also by the genetic background or ecotype of the mushroom species (Bazzicalupo et al. 2020; Sácký et al. 2022). In light of the fact that the consumption of heavy metal-contaminated mushrooms can adversely affect human health, in recent years considerable attention has been paid to the assessment of health risks associated with mushroom consumption. Moreover, several studies have ranked heavy metal contamination among the main risk factors affecting the safety of edible mushrooms (Jia et al. 2016; Han et al. 2020; Qiu et al. 2024).

Cadmium (Cd) is considered to be one of the most toxic heavy metals. Numerous studies have confirmed that consumption of Cd-contaminated foodstuffs, even at low levels, adversely affect kidneys, liver, bone and heart, and in extreme cases can cause death (Genchi et al. 2020; Charkiewicz et al. 2023). Unfortunately, mushrooms easily absorb Cd from the substrate in which they grow, indicating that mushroom substrate contaminated with Cd poses a serious threat to mushroom production, safety and quality (Širić et al. 2022). It is accordingly obvious that mushroom cultivation in Cd-contaminated substrates should be avoided.

The Cd concentrations found in agricultural wastes suitable for cultivating oyster mushrooms typically ranges from 0.05 mg·kg^{-1} and 0.40 mg·kg^{-1} of dry mass (Stoknes et al. 2019). However, agricultural waste/residues mainly used for mushroom cultivation can have considerably higher Cd concentrations, especially if the plant material originates from agricultural areas with intensive use of agrochemicals and inorganic fertilisers that contain Cd and other heavy metals (Mubeen et al. 2023). When viewed in this light, it is clear that mushroom growth substrate is a crucial factor affecting the quality and safety of edible mushrooms.

Recognising this specificity, as well as the fact that pink oyster mushrooms are becoming one of the most popular mushrooms, the aim of this study was to evaluate the ability of pink oyster mushrooms to absorb Cd from substrates contaminated with Cd at concentrations of 0, 20, 50 and 100 mg·kg^{-1} and to assess the human health risks associated with the consumption of these mushrooms. We hypothesised that increasing Cd concentration in mushroom substrate will result in increased Cd concentration in mushroom fruiting bodies. We also hypothesised that mushrooms grown in cadmium-contaminated substrates would pose a risk to human health from the Cd contamination point of view.

MATERIAL AND METHODS

Experimental design. This study was carried out from April 2024 to June 2024 in a mushroom growth chamber at the Faculty of Agriculture and Food Sciences, University of Sarajevo. An experiment was carried out in a completely randomised design and included four treatments (four Cd contamination levels i.e. 0, 20, 50 and 100 mg·kg^{-1}) with three replications per treatment.

Substrate preparation, inoculation and incubation. Mixture of beech sawdust and wheat bran (in a ratio of 80 : 20) containing 0, 20, 50 and 100 mg·kg^{-1} of Cd, was used as a substrate in this study in order to assess the ability of pink oyster mushrooms to absorb Cd from growing medium. Substrate was prepared as follows: 10 kg of beech sawdust (granulation from 0.5 mm to 1 mm) and 2 kg of wheat bran were transferred to a styrofoam container and mixed well. After mixing, 25 L of water was added and the substrate components

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were mixed again until there was no more water at the bottom of the container (the moisture content was between 60–65%). Thereafter, the substrate was divided into four equal parts and then each part was spiked with an aqueous stock solution of $CdSO_4$ to result in final Cd concentrations of 0, 20, 50 and 100 mg·kg^{-1} fresh weight, respectively. To ensure uniform distribution of Cd, the solution of $CdSO_4$ (500 mL) was sprayed onto the substrate, which was thoroughly mixed during spraying. Following this, sawdust-wheat bran substrates contaminated with different concentrations of Cd were transferred to individual plastic grow boxes (1 kg of substrate was placed in each plastic box) and then pasteurised at 65 °C for 8 h. A total of 12 plastic boxes were used in this study (four treatments i.e. with three replications).

Following pasteurisation, plastic boxes were cooled at room temperature, and then inoculated with pink oyster mushroom mother spawn at a concentration of 10%. Mushroom mother spawn was obtained from Urban Farm Mikić (Orašje, Bosnia and Herzegovina), and was prepared using sorghum grains as substrate in accordance with the method of spawn preparation described by Stamets and Chilton (1983).

Following inoculation, the plastic boxes were transferred to a mushroom growth chamber and incubated at 24 ± 2 °C in darkness with relative humidity of 80% for 2–3 weeks, by which time the mushroom mycelium fully colonised the substrate. Thereafter, the plastic box lids were cut (holes of 1 cm in diameter) at six positions, and the temperature in a climate chamber was decreased to 16 °C for further three days to stimulate fructification. Plastic boxes in a climate chamber were then exposed to daylight for 12 h per day (on/off cycle) at 18 ± 2 °C and 85% relative humidity until harvest.

Substrate chemical analysis. A chemical analysis of the substrate samples took place after its preparation and before it was contaminated with Cd. To prepare substrate samples for chemical analysis, the following procedure was followed: mushroom substrate samples were dried in an oven at 40 °C until they achieved a constant weight, then ground into a fine powder with a mortar and pestle. The resulting powder was passed through a 2 mm sieve and subsequently analysed in the laboratory to evaluate basic chemical properties, including substrate pH, organic matter content, levels of available forms of phosphorus (P_2O_5) and potassium (K_2O), and the contents of Cd, Cu, Zn, Mn, Ni, Cr and Pb.

The pH of substrate was determined with a pH meter in distilled water (pH H_2O) and in a 1 mol·L^{-1} KCl solution (pH KCl) at a ratio of 1 : 5 (ISO 10390, 2005). The organic matter content was measured using the H_2SO_4-$K_2Cr_2O_7$ oxidation method (ISO 14235, 1998), while the available forms of phosphorus and potassium were evaluated using the ammonium lactate method (Egnér et al. 1960).

Extraction of heavy metals from substrate sample was performed by mixing 3 g of the soil with 21 mL aqua regia solution (a mixture of concentrated HNO_3 and HCl in a ratio of 1 : 3). This mixture was allowed to react for 16 h (overnight) in a fume hood. Following this, the mixture was heated to its boiling point on a hotplate under reflux for 2 h. After cooling, the mixture was filtered through quantitative filter paper into a 100 mL flask, which was subsequently filled to the mark with deionised water (ISO 11466, 1995).

Heavy metals contents in the extracts were determined using atomic absorption spectrometry (ISO 11047, 1998) with a Shimadzu AA-7000 spectrophotometer (Tokyo, Japan). Calibration standards were prepared from standard stock reagents supplied by Merck (Darmstadt, Germany), which contained 1 000 mg·kg^{-1} of the heavy metals analysed.

Mushroom harvesting, observation and measurement. Pink oyster mushrooms were harvested when the edges of the mushrooms started to crack and split. Three flushes were harvested during total cropping period and interval between flushes varied from five to seven days. Yield from each flush and total yield were recorded in weight (g).

The biological efficiency (*BE*) of pink oyster mushroom was calculated using the equation outlined by Thongsook and Kongbangkerd (2011):

$$BE = \frac{\text{fresh mass of harvested mushroom}\,(\text{g})}{\text{substrate dry mass}\,(\text{g})} \times 100 \qquad (1)$$

Preparation and chemical analysis of mushroom samples. Samples of complete fruiting bodies from each plastic box were dried by an oven at 60 °C to a constant weight. Following this, the dried mushroom fruiting bodies were ground into a fine powder using an electric blender and stored in paper bags until later use.

Extraction of heavy metals from mushroom sample was done using a mixture of HNO_3 and H_2SO_4 in a ratio of 2.5 : 1 (12 mL of mixture for 1 g of dry sample). This mixture was heated up to 80 °C on a hot plate for 3 h and then cooled to room temperature. After that, the mixture was filtered through quantitative filter paper in a 25 mL flask and diluted with deionised water to the mark (Lisjak et al. 2009).

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Heavy metal concentrations (Cd, Cu, Zn, Mn, Ni, Cr and Pb) in mushroom samples were determined by atomic absorption spectrophotometry using the Shimadzu AA-7000 device (Shimadzu Instruments, Tokio, Japan). Working solutions for each investigated heavy metal were prepared by diluting the standard stock solutions (Merck, Germany) with deionised water as necessary.

Health risk analysis. The potential human health risk of Cd and some other heavy metals via consumption of pink oyster mushrooms was assessed using target hazard quotient (*THQ*), which was described in detail by the United States Environmental Protection Agency (USEPA 2011). In this study, the *THQ* values for each heavy metal found in pink oyster mushrooms was calculated using the following equation (USEPA 2011):

$$THQ = \frac{c \times IR \times ER \times \mathrm{ED}}{ET \times BW \times RfD} \quad (2)$$

where: c – concentration of contaminant in analysed mushroom sample (mg·kg^{-1}); IR – food (mushroom) ingestion rate (0.0066 kg·person^{-1}·day^{-1}); EF – exposure frequency (365 days); ED – exposure duration for adult (70 years); ET – averaged exposure time (25 550 days), BW – body weight (for adults 70 kg, for children 32 kg); RfD – the oral reference dose i.e. the highest level of contaminant at which no adverse health effects are expected.

According to USEPA (2011) the RfD values for Pb, Cd, Cu, Zn, Mn, Ni and Cr are 0.004, 0.001, 0.040, 0.300, 0.140, 0.020 and 0.003 mg·kg^{-1}·day^{-1}, respectively.

Any *THQ* value greater than 1 indicates a high risk of non-cancerous diseases due to consumption of contamination food.

Statistical analysis. All data were performed using SAS 9.4 (SAS Institute, Cary, NC, USA) software. The least significant difference (LSD) test at a 5% probability level was used to compare the treatment's means.

RESULTS

Results of the basic chemical analysis of the mushroom growing media i.e. substrate before Cd-contamination are presented in Table 1. All results are expressed on a dry mass basis.

The mushroom substrate used in this study was slightly acidic and had very high levels of organic matter and available forms of potassium and phosphorus. The level of Cd, Cu, Zn, Ni, Cr and Pb in mushroom substrate was found to be lower than permissible limits recommended by legislation in Bosnia and Herzegovina (OF FBiH 2009). The safe limits for Cd, Cu, Zn, Ni, Cr and Pb in agricultural soils recommended by legislation in Bosnia and Herzegovina are 1.5, 80, 100, 40, 100 and 100 mg·kg^{-1} dry mass, respectively. The levels of investigated heavy metals (Cd, Cu, Zn, Mn, Ni and Cr) in the pink oyster mushroom fruiting bodies grown on substrates contaminated with Cd are shown in Table 2. All results are expressed on a dry mass basis in mg·kg^{-1}.

Table 1. Basic chemical characteristics of the mushroom substrate

Chemical properties	Unit	Value
pH (H_2O)	pH unit	6.9
pH (KCl)	pH unit	6.4
organic matter	%	78.7
P_2O_5	mg·(100 g)$^{-1}$	80
K_2O	mg·(100 g)$^{-1}$	230
Cd	mg·kg^{-1}	0.25
Cu	mg·kg^{-1}	1.9
Zn	mg·kg^{-1}	15.4
Mn	mg·kg^{-1}	62.8
Ni	mg·kg^{-1}	0.4
Cr	mg·kg^{-1}	1.3
Pb	mg·kg^{-1}	n.d.

n.d. – not detected

The concentrations of Cu, Zn, Mn, Ni and Cr in pink oyster mushrooms were below their respective maximum permissible levels prescribed by World Health Organization (WHO 1989). Accordingly, the maximum permissible limits for Cu, Zn, Mn, Ni and Cr in vegetables/mushrooms are 73, 100, 500, 67 and 2.3 mg·kg^{-1}, respectively. From this point of view, consumption of mushrooms from the studied site could be considered as safe for human health. However, on the other hand, the concentration of Cd in mushrooms grown on substrates contaminated with Cd was much higher than the permissible level recommended by WHO of 0.1 mg·kg^{-1}, which was expected.

The potential health risks associated with food consumption can be predicted using a target hazard quotient (*THQ*). The Table 3 summarises the established *THQ* values for each heavy metal found in pink oyster mushrooms.

As expected, the *THQ* values in analysed pink oyster mushroom samples were highest for Cd. The *THQ* values of Cd for adults and children in all mushrooms grown on Cd-contaminated substrates were great-

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Table 2. Heavy metal levels (mg·kg^{-1} dry mass) in pink oyster mushrooms grown on Cd-contaminated substrates

Treatments*	Cd	Cu	Zn	Mn	Ni	Cr
1 (control)	1.8 ± 0.4^{d**}	6.8 ± 2.1	41.2 ± 3.6	16.9 ± 3.1	0.21 ± 0.08	0.91 ± 0.18
2 (20 mg·kg^{-1})	11.1 ± 3.7^{c}	5.9 ± 2.4	40.6 ± 2.8	21.8 ± 5.4	0.15 ± 0.09	1.03 ± 0.22
3 (50 mg·kg^{-1})	18.2 ± 1.5^{b}	6.9 ± 2.6	41.3 ± 1.9	16.8 ± 4.9	0.13 ± 0.08	0.91 ± 0.12
4 (100 mg·kg^{-1})	23.8 ± 3.1^{a}	8.1 ± 3.7	41.5 ± 2.4	21.5 ± 5.2	0.18 ± 0.07	1.07 ± 0.19
$LSD_{0.05}$	1.94	–	–	–	–	–

*treatments: 1 – substrate without Cd contamination (control), 2 – substrate contaminated with Cd (20 mg·kg^{-1}), 3 – substrate contaminated with Cd (50 mg·kg^{-1}), 4 – substrate contaminated with Cd (100 mg·kg^{-1}); **averages denoted by the same letter in the same column indicate no significant difference ($P < 0.05$); $LSD_{0.05}$ – least significant difference test at a 5% probability level

Table 3. Heavy metal *THQ* values due to consumption of pink oyster mushrooms

Treatments*	Cd		Cu		Zn		Mn		Ni		Cr	
	adult	child	adult	child	adult	child	adult	child	adult	child	adult	child
1 (control)	0.17	0.37	0.02	0.04	0.01	0.03	0.01	0.02	0.001	0.002	0.03	0.06
2 (20 mg·kg^{-1})	1.04	2.29	0.01	0.03	0.01	0.03	0.01	0.03	0.001	0.002	0.03	0.07
3 (50 mg·kg^{-1})	1.71	3.75	0.02	0.04	0.01	0.03	0.01	0.02	0.001	0.002	0.03	0.06
4 (100 mg·kg^{-1})	2.24	4.91	0.02	0.04	0.01	0.03	0.01	0.03	0.001	0.002	0.03	0.07

*treatments: 1 – substrate without Cd contamination (control), 2 – substrate contaminated with Cd (20 mg·kg^{-1}), 3 – substrate contaminated with Cd (50 mg·kg^{-1}), 4 – substrate contaminated with Cd (100 mg·kg^{-1}); *THQ* – target hazard quotient

er than 1, suggesting that long-term consumption of these mushrooms can lead to acute toxicity and diseases from a Cd point of view. The *THQ* values of Cu, Zn, Mn, Ni and Cr were much less than 1.

The yield and biological efficiency values of pink oyster mushrooms grown on substrates contaminated with Cd at concentrations of 0, 20, 50 and 100 mg·kg^{-1} are given in Table 4.

As shown in Table 4, the highest yield and biological efficiency values of pink oyster mushrooms were determined in substrate 1 (non-contaminated substrate) whereas the lowest were found in substrate 4 (substrate contaminated with Cd at concentrations of 100 mg·kg^{-1}).

DISCUSSION

The current study showed that pink oyster mushrooms collected from Cd-contaminated substrates accumulate Cd at high concentrations compared to the maximum acceptable limit set by WHO (1989). Accordingly, the maximum permissible limit for Cd

Table 4. Yield performance of pink oyster mushroom grown on Cd-contaminated substrates

Treatments*	Fruiting bodies				Biological efficiency (%)
	I flush (g·packet^{-1})	II flush (g·packet^{-1})	III flush (g·packet^{-1})	total yield (g·packet^{-1})	
1 (control)	61.1 ± 8.7	43.0 ± 6.1	17.0 ± 3.3	121.1 ± 8.5^{a**}	35.6 ± 4.5^{a}
2 (20 mg·kg^{-1})	57.2 ± 7.1	26.5 ± 4.2	14.1 ± 5.3	97.5 ± 7.3^{b}	28.7 ± 3.0^{b}
3 (50 mg·kg^{-1})	63.3 ± 9.1	12.2 ± 4.1	–	75.5 ± 5.4^{c}	21.6 ± 3.4^{c}
4 (100 mg·kg^{-1})	12.4 ± 2.8	8.1 ± 2.3	–	20.5 ± 3.7^{d}	6.0 ± 4.9^{d}
$LSD_{0.05}$	–	–	–	6.4	4.1

*treatments: 1 – substrate without Cd contamination (control), 2 – substrate contaminated with Cd (20 mg kg^{-1}), 3 – substrate contaminated with Cd (50 mg·kg^{-1}), 4 – substrate contaminated with Cd (100 mg·kg^{-1}); **averages denoted by the same letter in the same column indicate no significant difference ($P < 0.05$); $LSD_{0.05}$ – least significant difference test at a 5% probability level

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is 0.1 mg·kg^{-1}. It is interesting that in this study, the concentration of Cd in pink oyster mushrooms grown on non-contaminated substrates was also higher than previous mentioned permissible level, indicating that these mushrooms have a high ability to absorb Cd from substrates in which they grow.

The study results also showed that the Cd accumulation in pink oyster mushrooms increased sharply with increasing Cd content in mushroom growing substrates and ranged from 1.8 mg·kg^{-1} (in a non-contaminated substrate) to 23.8 mg·kg^{-1} dry mass (in substrate contaminated with 100 mg·kg^{-1}). These findings support the hypothesis that the levels of Cd in the mushroom fruiting bodies strongly depend on the Cd content of the substrate in which they grow, and this hypothesis has been confirmed by a number of studies (Muszyńska et al. 2018; Andronikov et al. 2023). This fact also leads to the conclusion that pink oyster mushrooms do not possess highly effective mechanisms to block or avoid the uptake of Cd from growing medium. From a health point of view, this is highly undesirable because the Cd exposure increased the risk of liver dysfunction, cardiovascular disease, atherosclerosis and brain damage (Naija and Yalcin 2023). Target hazard quotient (*THQ*) values for Cd obtained in this study support this observation. Namely, the pink oyster mushrooms grown on Cd-contaminated substrates had a *THQ* value for Cd greater than 1, indicating a potential health risk to humans.

Although mushrooms do not possess highly effective mechanisms to block or avoid the uptake of Cd from growing medium, it is important to note that they have developed various mechanisms to neutralise Cd within their body, including, among others, intracellular sequestration and extracellular complexation (Subašić et al. 2022).

However, the ability of mushrooms to neutralise Cd within their body is not unlimited, which means that if mushrooms are grown in a medium highly contaminated with Cd, it is realistic to expect that Cd negatively affects the mushroom growth and development. The results of this study related to mushroom yield parameters confirm this observation. Accordingly, as shown in Table 4, a consistent decrease in pink oyster mushrooms yield and biological efficiency was observed with increasing Cd concentration in growing substrate. Moreover, substrates highly contaminated with Cd (substrate 3 and 4) did not produce any growth beyond the third flush. These results indicate that elevated concentrations of Cd in the growing medium suppress fruiting body induction in pink oyster mushrooms.

In this study, *THQ* values in analysed mushroom samples for Cu, Zn, Mn, Ni and Cr were much less than 1 for both adults and children, suggesting that these heavy metals do not pose a potential health risk for consumers through the consumption of pink oyster mushrooms from the studied site. However, the obtained *THQ* values for Cu, Zn, Mn, Ni and Cr were expected to be at low level since the content of these heavy metals in growing medium for mushrooms were below the permissible limits prescribed by WHO (1989).

Interestingly, in this study, the levels of Zn and Cu in pink oyster mushrooms were much higher than in the substrate in which they were grown. These findings indicate that pink oyster mushrooms have the capability to transfer not only Cd, but also Zn and Cu from growing medium to their fruiting bodies very efficiently. Similar findings have been reported by Golian et al. (2021). From the point of view of Cu and Zn, this was not unexpected, considering that Zn and Cu are involved in various metabolic processes in mushrooms (Mirończuk-Chodakowska et al. 2019).

Although fungi possess specific transport systems for the uptake of manganese, such as transporters from the NRAMP (Natural Resistance-Associated Macrophage Protein) family, the levels of Mn in pink oyster mushrooms grown on Cd-contaminated substrates were lower than in the initial substrate. Numerous studies have also shown that the ability of mushrooms to accumulate Mn from the growing substrate is relatively low compared to Cu and Zn (Falandysz and Frankowska 2007; Podlasińska et al. 2015). It is interesting that plants, unlike mushrooms, can accumulate large amounts of Mn in their above-ground parts. Mn is an essential mineral element in plants, which plays a key role in various physiological processes, particularly photosynthesis, and therefore plants tend to absorb Mn as much as possible from growing medium (Alejandro et al. 2020). Contrastingly, mushrooms do not perform photosynthesis, which results in a lower requirement for manganese. This is probably the reason why Mn accumulation in fungi is less compared to that in plants.

In this study, Cr levels were fairly similar in the pink oyster mushrooms and in the substrate in which they were grown, suggesting that pink oyster mushrooms have the ability to transfer Cr from the substrate to their fruiting bodies very efficiently. From the consumer's point of view, this finding is consid-

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ered to be undesirable because of adverse effects of Cr on human health.

CONCLUSION

The results of the present study suggest that pink oyster mushrooms have the capability to absorb Cd from the substrate in which they grow. In addition, the obtained results for *THQ* of Cd point to the conclusion that the consumption of pink oyster mushrooms cultivated in Cd-contaminated substrates could produce negative health effects. From the standpoint of Cd toxicity, it should be noted that consuming pink oyster mushrooms may entail a potential health risk, even when they are grown on non-contaminated substrates, due to their ability to accumulate Cd. Study results also showed that the substrate contaminated with Cd strongly affects mushrooms growth and development and thus reduces their yield.

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