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HEALTH RISK ASSESSMENT OF CONSUMING WILD EDIBLE MUSHROOMS (*M. PROCERA* AND *C. CIBARIUS*) BASED ON TOTAL MERCURY CONTENT

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ABSTRACT

The consumption of wild edible mushrooms is common in Central Europe; however, their ability to accumulate mercury (Hg) may pose health risks. This study evaluated total mercury content in the caps and stipes of *Macrolepiota procera* and *Cantharellus cibarius* collected from 34 localities in Slovakia, including historically contaminated mining areas. In total, 148 mushroom samples and corresponding soil samples were analyzed using AMA-254 analyzer. Soil contamination was assessed using the contamination factor (C_f), while mercury uptake and distribution were evaluated using the bioaccumulation factor (BAF) and translocation factor ($Q_{c/s}$). Potential health risks were determined using provisional tolerable weekly intake (%PTWI) and target hazard quotient (THQ). Soil mercury concentrations ranged from 0.04 to 7.39 mg·kg^{-1}, with the highest values recorded in former mining regions. *M. procera* showed significantly higher mercury levels compared to *C. cibarius*. In both species, mercury concentrations were higher in caps than in stipes. BAF values confirmed *M. procera* as a mercury accumulator, whereas *C. cibarius* exhibited lower accumulation capacity. Health risk assessment revealed that %PTWI frequently exceeded 100% for *M. procera*, particularly in contaminated areas, and THQ values above 1 indicated potential non-carcinogenic health risks associated with long-term consumption. These findings emphasize the need for monitoring mercury contamination in wild mushrooms and highlight potential risks related to their consumption from environmentally burdened sites.

Keywords: Mercury, Edible mushroom, Risk assessment, Contamination

INTRODUCTION

The consumption of wild edible mushrooms represents a deeply rooted gastronomic tradition in many countries, particularly in Central Europe (**Árvay *et al.*, 2022**). Mushrooms is considered beneficial to health because they are a rich source of biologically active compounds, including high-quality proteins, dietary fiber, B-group vitamins (such as riboflavin, niacin, and pantothenic acid), and essential minerals like potassium, phosphorus, and selenium. In addition, mushrooms contain various bioactive compounds, such as polysaccharides (particularly β-glucans), phenolic compounds, and antioxidants, which may contribute to supporting the immune system, reducing oxidative stress, and promoting overall health (**Valverde *et al.*, 2015**; **Kalač, 2013**). Despite their nutritional benefits, however, mushrooms possess a specific physiology that enables them to efficiently absorb and accumulate risk elements from the surrounding environment, even in areas not considered extremely polluted (**Procházka *et al.*, 2023**; **Širić *et al.*, 2022**; **Melgar *et al.*, 2009**). One of the most critical contaminants is mercury (Hg), which the World Health Organization ranks among the ten chemicals of greatest public health concern (**WHO 2024**). In addition, mercury has the ability to accumulate in living organisms and enter the food chain through bioaccumulation processes and biomagnification (Guédron and Acha, 2021; **Sunderland, 2007**).

The health risks associated with mercury exposure are complex and significant. Long-term intake of mercury, even at relatively low concentrations, can result in toxic effects on multiple organ systems. In particular, mercury exposure has been shown to damage the nervous, digestive, and immune systems, while also adversely affecting the lungs and kidneys (**WHO 2024; Ab Rahman, 2022**; **Rzymski *et al.*, 2015**). Furthermore, chronic exposure has been associated with cardiovascular dysfunction, including hypertension and impaired endothelial function (**Azevedo *et al.*, 2016**). Mercury toxicity has also been linked to reproductive effects, such as reduced sperm quality and potential impacts on fertility and pregnancy outcomes (**Kumar *et al.*, 2024b**). Overall, due to its ability to accumulate in biological tissues and disrupt essential cellular processes, mercury represents a significant threat to human health even at relatively low levels of exposure (**Bernhoft 2012**).

In the context of Slovakia, this risk is amplified by the presence of numerous environmental burdens and historical mining districts, where mercury concentrations in soils frequently exceed legal limits (IS EZ 2026). Former mining and smelting activities, particularly in regions such as Central Spiš or historical mercury deposit in Malachov, have resulted in long-term contamination of soils, sediments, water bodies and biota (**Navrátil *et al.*, 2026**; **Andráš *et al.*, 2021; Demková *et al.*, 2019**). These legacy contaminations represent a long-term environmental burden because mercury is highly stable in the environment and can remain available for biological uptake for decades after mining activities cease (**Kimáková *et al.*, 2017**; **Árvay *et al.*, 2014**).

Wild-growing mushrooms are particularly important bioindicators of heavy metal contamination due to their ability to absorb and accumulate metals directly from the soil substrate through extensive mycelial networks (**Ali *et al.*, 2024**; **Qin at al., 2024**).Numerous studies demonstrate that mercury is unevenly distributed within mushroom fruiting bodies, with the highest concentrations typically detected in the caps (pileus) and spore-bearing tissues rather than in the stipes (**Falandysz and Gucia, 2008**). As a consequence, the edible parts most frequently consumed by humans may contain substantially higher concentrations of mercury than the surrounding substrate itself (**Campos *et al.*, 2009**).

In areas affected by historical mining activities, the consumption of contaminated wild mushrooms (and vegetables) may lead to mercury intake levels exceeding recommended safety limits by several hundred percent, especially in populations with high mushroom consumption rates (**Chen *et al.*, 2024**; **Kimaková *et al.*, 2017; Tomáš *et al.*, 2012**). According to risk assessment studies, such exposure may pose a significant risk of non-carcinogenic health effects, particularly for vulnerable groups such as children due to their lower body weight and higher relative dietary intake (**Árvay *et al.*, 2021**).

The aim of this study was to evaluate the mercury content in the caps and stipes of two wild-growing mushroom species *M. procera* and *C. cibarius*, in order to assess their potential to accumulate mercury from contaminated environments. Specifically, the study focused on (i) determining the concentration of mercury in the soil from the studied localities, (ii) evaluating the relationship between soil contamination and mercury accumulation in mushroom fruiting bodies with respect to their bioaccumulation capacity, and (iii) assessing the potential health risks associated with the consumption of the analysed mushroom species using selected health risk assessment indicators. In addition, the study aimed to compare the mercury accumulation patterns between the two mushroom species investigated.

MATERIAL AND METHODS

Mushroom and soil sampling and analysis

Two edible mushroom species, *Macrolepiota procera* (121 fruiting bodies) and *Cantharellus cibarius* (158 fruiting bodies), were collected between 2020 and 2024 at selected sites in Slovakia (n = 34) (Figure 1). For analytical purposes, individual fruiting bodies were not treated as separate samples. Instead, a sample was defined as a group of at least three fruiting bodies of the same species collected at a single sampling site. Based on this criterion, a total of 148 mushroom samples were prepared and analyzed.Sampling was conducted opportunistically at sites where the target mushroom species were present and environmental conditions were suitable for fruiting body development.. Soil samples (100 g, 0.00–0.10 m) were collected following the same sampling design, with each mushroom sample being paired with a corresponding soil sample taken from the immediate vicinity of the mushroom collection site. The mushroom samples were cleaned of impurities, leaves, and other debris and stored in ventilated polyethylene boxes. On the same day, under laboratory conditions, the mushrooms were rinsed with deionized water; caps were separated from stipes, and both parts were sliced and dried in a laboratory drying oven with forced air circulation (Memmert UF 110 m, Memmert GmbH & Co. KG, Schwabach, Germany) at 40 °C for approximately 22 hours. The dried mushroom samples were homogenized using a rotary homogenizer (IKA A 10 basic, IKA-Werke GmbH & Co. KG, Staufen, Germany) and stored in sealable polyethylene bags until analysis. Soil samples were cleaned of roots and debris and then air-dried under laboratory conditions for three weeks, homogenized, sieved through a 2 mm mesh, and stored in paper bags until analysis. Mercury (Hg) concentrations in mushrooms and soil were determined by cold vapor atomic absorption spectrometry (CV-AAS) using an AMA-254 analyzer (AlTec spol. s r. o., Prague, Czech Republic) equipped with an ASS-254 automatic solid sampler (AlTec spol. s r. o., Prague, Czech Republic). Quantitative determination of Hg was performed at λ = 253.7 nm. The detection limit for mercury was 0.0011 mg·kg^{-1} DW and the limit of quantification was 0.0031 mg·kg^{-1} DW (**Száková *et al.*, 2003**). For quality control, two certified reference materials (CRMs) were analyzed: ERM-CC 141 (loamy soil; IRMM Geel, Belgium) and ERM-CE 278k (mussel tissue; IRMM Geel, Belgium). Method accuracy was evaluated using the certified reference materials ERM-CC141 and ERM-CE278k. Recoveries of 99.1% and 99.8%, respectively, were achieved, indicating excellent analytical accuracy. Precision was determined from six independent replicate measurements of each CRM and expressed as relative standard deviation (RSD). The RSD values were lower than 3% for both reference materials, confirming the high precision and repeatability of the AMA-254 analytical method for total mercury determination.

Mushroom samples

The contamination factor (C_f) was used to determine the level of mercury pollution in soils at individual sampling sites (**Hakanson, 1980**), and calculated according to the following equation:

$$C_f = C_{o\text{–}1i} / C_{ni} \qquad (1)$$

where: $C_{o\text{–}1i}$ represents the Hg content in soil samples and C_{ni} is the reference (background) value of mercury in soils (set at 0.08 mg·kg^{-1} DW according **Šefčík *et al.* (2008)**. Based on **Hakanson (2008)**, C_f values were classified into four categories: (i) low contamination factor (C_f<1), (ii) moderate contamination factor (1≤C_f<3), (iii) considerable contamination factor (3≤C_f<6), and (iv) very high contamination factor (C_f≥6)
To assess the ability of mushrooms to uptake mercury from soil into their aboveground parts, the bioaccumulation factor (BAF) was determined. The bioaccumulation factor of the studied mushroom species was calculated as follows:

$$BAF = Hg_m / Hg_s \qquad (2)$$

where: Hg_m is the mercury concentration in mushrooms and Hg_s is the mercury concentration in soil (mg·kg^{-1} DW). BAF was calculated separately for individual parts of the fruiting bodies (caps and stipes). *BAF* results were classified into three categories: *BAF*<1 indicates mercury-excluding (excluder) species, BAF>1 indicates mercury-accumulating (accumulator) species, and BAF=1 indicates indicator species (**Jonczak *et al.*, 2021; Dryżałowska and Falandysz, 2014**).To evaluate the translocation factor ($Q_{c/s}$) expresses the ratio of mercury concentration in the cap to that in the stipe and was calculated using the following equation:

$$Q_{c/s} = \mathrm{Hg_{cap}} / \mathrm{Hg_{stipe}} \qquad (3)$$

where: $\mathrm{Hg_{cap}}$ is the mercury concentration in mushroom caps and Hg_{stipe} is the mercury concentration in mushroom stipes. To evaluate the potential risk arising from long-term consumption of edible mushrooms, the provisional tolerable weekly intake (PTWI) was used. According to the **WHO (2011)**, the *PTWI* for mercury was set at 0.004 mg·kg^{-1} body weight (BW), corresponding to 0.28 mg per adult. The percentage of *PTWI* (*%PTWI*) was calculated using the following equation:

$$\%PTWI = (\text{Hg in mushroom} \times \text{intake}) / \text{PTWI(Hg)} \times 100 \qquad (4)$$

where Hg in mushroom is the mercury concentration (mg·kg^{-1} fresh weight (FW)) in the mushroom fruiting body, intake represents the consumption of the studied mushrooms (kg·week^{-1} FW), and *PTWI(Hg)* = 0.28 mg per adult person. Values exceeding 100% may be considered potentially hazardous. Fresh weight of mushrooms was calculated assuming that dry matter constituted 10% of the mushroom fruiting body (**Kalač, 2010**). According to the **Statistical Office of the Slovak Republic (2018)**, the average consumption of the category "*other vegetables and mushrooms*" was set at 0.23 kg fresh weight per week.
To evaluate the potential health risks associated with long-term mushroom consumption, the target hazard quotient (THQ) was applied. THQ represents the ratio between the estimated exposure to a toxic element and the maximum reference dose at which no harmful effects on human health are anticipated. The THQ was calculated using the following equation:

$$THQ = ((Efr \times ED \times ADC \times CE) / (RfDo \times BW \times ATn)) \times 10^{-3} \qquad (5)$$

where: *Efr* denotes exposure frequency (365 days), *ED* is the exposure duration (70 years), *ADC* represents the average daily intake of fresh mushrooms (33 g·day^{-1}) (**Statistical Organization of Slovak Republic, 2021**), and CE is the mean mercury concentration in mushroom samples (mg·kg^{-1} FW). *RfDo* corresponds to the oral reference dose for mercury (0.0003 mg·kg^{-1}·day^{-1}) (**Kalač 2019**). *BW* refers to the average body weight (70 kg), while *ATn* is the average exposure period (365 days × 70 years = 25,550 days). The factor 10^{-3} accounts for unit conversion. *THQ* values below 1 indicate that non-carcinogenic health risks are unlikely, whereas values exceeding 1 suggest a potential risk of adverse health effects.
A limitation of the present health risk assessment is that the calculations were based on the average mushroom consumption rate reported for the Slovak population. Since actual consumption may differ considerably among individuals, especially among frequent mushroom consumers, the estimated %PTWI and THQ values should be regarded as indicative rather than absolute measures of health risk.

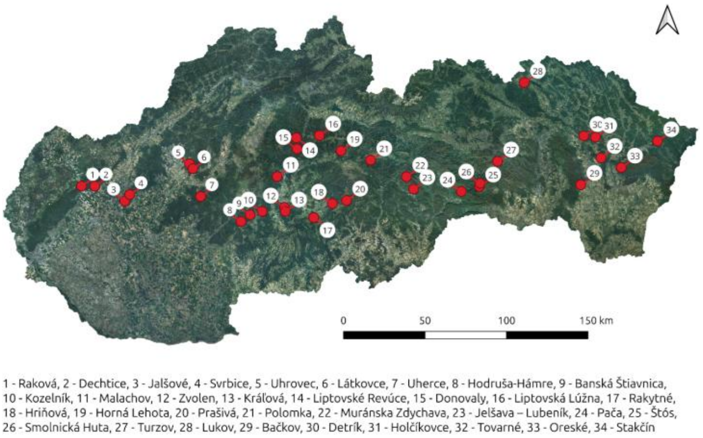

Figure 1 Edible mushroom and soil sampling sites

Data analysis

All statistical analysis were performed in program Past4.03 (**Hammer *et al.*, 2001**). Non-parametric Mann–Whitney U test was used to determine differences in individual evaluated parameters between the two assessed mushroom species.

RESULTS AND DISCUSSION

Total mercury content in the soils ranged from 0.04 to 7.388 mg·kg^{-1} . Limit value for mercury in Slovak soils was set to 0.5 mg·kg^{-1} (Act. No. 2020/2004 Coll. of Laws.). This value was exceeded at 5 sampling sites. The highest value was measured at the Malachov sampling site (7.39 mg·kg^{-1}), followed by the Štóske sedlo (2.48 mg·kg^{-1}), Smolnícka Huta (1.95 mg·kg^{-1}) and Turzov (1.59 mg·kg^{-1}) sites. The elevated soil Hg concentrations recorded at Malachov, Štóske sedlo, Smolnícka Huta and Turzov are consistent with the historical mining activities documented for these regions, moreover, all of them are classified as environmentally burdened localities. (IS EZ 2026). The results show that former mining areas remain important sources of environmental mercury contamination even decades after their active operation have ceased. High mercury values in the soil were also reflected in the values of the contamination factor, which are shown in Figure 2. Soil quality in former mining areas in Slovakia has long been influenced by high mercury concentrations, which are also present in other components of the environment, such as water resources and the bodies of living organisms (**Demková *et al.*, 2023**; **Dadová *et al.*, 2016**; **Singovszká *et al.*, 2016**). In numerous environmental studies, the transfer of toxic elements such as mercury from contaminated soil into plants via the root system – and their subsequent accumulation in plant tissues – has been monitored over long periods, allowing assessment of bioaccumulation and contaminant transfer within ecosystems (Chen *et al.*, 2024; Zhou *et al.*, 2021).

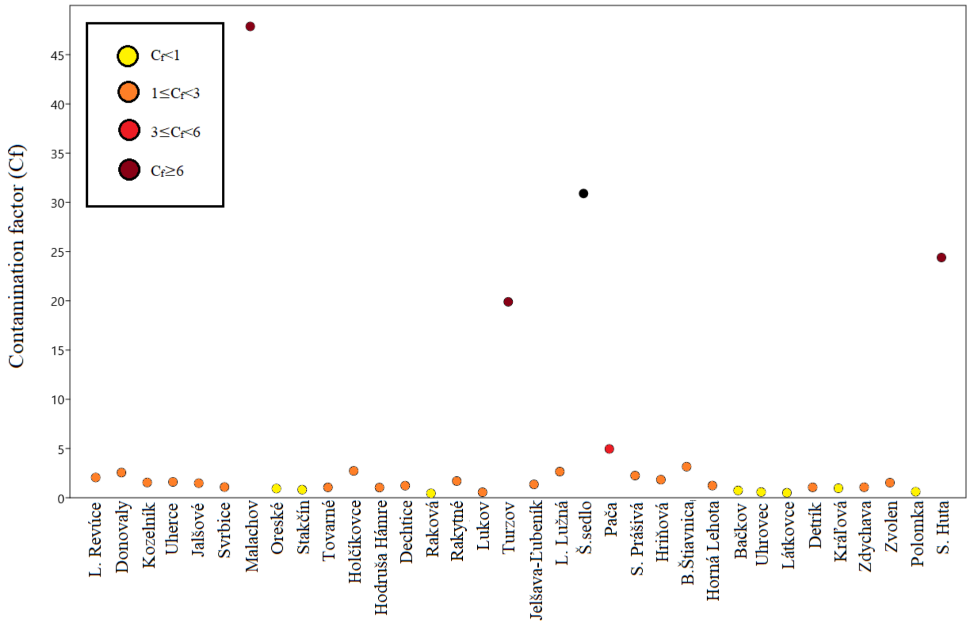

Figure 2 Average contamination factor values (C_f) at individual sampling sites.

The total mercury content determined in the caps and stipes of the evaluated mushroom species is presented in Figure 3. The mercury content in *M. procera* ranged between 0.49–16.0 mg·kg^{-1} in the caps and 0.27–12.0 mg·kg^{-1} in the stipes. In *C. cibarius*, mercury concentrations ranged between 0.013–1.51 mg·kg^{-1} in the caps and 0.014–1.77 mg·kg^{-1} in the stipes. The results of the non-parametric Mann–Whitney U test showed that the mercury content determined in *M. procera* was significantly higher comparing *C. cibarius* in both caps and stipes ($p < 0.001$). In both evaluated species, it was clearly confirmed that mercury content was significantly higher in the caps compared to the stipes ($p < 0.0001$) (Figure 3). Several studies reported that in many mushroom species, risk elements accumulate in higher concentrations in the cap than in the stipe (**Širic *et al.*, 2016**). However, this pattern is not universal and depends on the mushroom species, the type of metal, and environmental condition (**Soceanu *et al.*, 2024**). The markedly higher Hg concentrations observed in *M. procera* compared with *C. cibarius* suggest species-specific differences in mercury uptake and accumulation. Studies focusing on the bioaccumulation capacity of *M. procera* consistently indicate that this species has a high ability to accumulate risk elements, particularly mercury (**Jančo *et al.*, 2021**). The detected Hg concentrations in fruiting bodies often reach markedly elevated levels, with higher contents generally observed in caps compared to stipes (**Falandysz and Gucia, 2008**). These findings point to potential health risks associated with regular consumption, especially when the mushrooms are collected from environmentally burdened or former mining areas (**Barea-Sepúlveda *et al.*, 2022**). Long-term exposure of mercury to the human body can result in toxic effects on the immune, digestive, and nervous systems, as well as on the eyes and skin, and may cause disorders of the respiratory, urinary, and reproductive systems (**WHO, 2026**; **Ab Rahman, 2022**; **Rzymski *et al.*, 2015**).

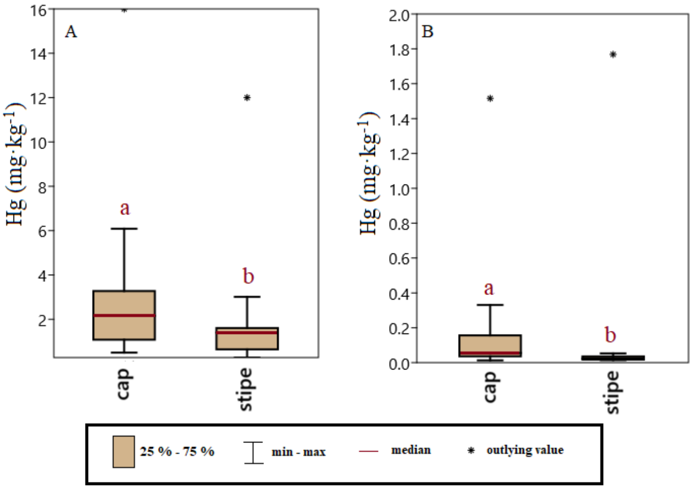

Figure 3 Mercury content in parts of a) *M. procera* and b) *C. cibarius* and the result of a non-parametric Mann-Whitney test (identical letters (a, b) indicate no significant differences in Hg content, whereas different letters indicate statistically significant differences in Hg conent between the cap and stipe (p<0.05))of a particular mushroom species

To further evaluate the relationship between mercury concentrations in soil and mushroom fruiting bodies, Spearman's rank correlation analysis was performed. In the case of *Macrolepiota procera*, significant positive correlations were found between mercury concentrations in soil and caps ($r = 0.43$, $p < 0.001$), as well as between mercury concentrations in soil and stipes ($r = 0.47$, $p < 0.001$). In addition, a very strong positive correlation was observed between mercury concentrations in caps and stipes ($r = 0.95$, $p < 0.001$), indicating a close association between mercury accumulation in different parts of the fruiting body (Figure 4). Similarly, in *Cantharellus cibarius*, mercury concentrations in soil were significantly positively correlated with mercury concentrations in caps ($r = 0.51$, $p < 0.001$) and stipes ($r = 0.41$, $p < 0.01$). A significant positive correlation was also found between mercury concentrations in caps and stipes ($r = 0.43$, $p < 0.01$). These results confirm that mercury concentrations in mushroom fruiting bodies are influenced by mercury levels in the underlying soil and support the assumption that soil represents an important source of mercury uptake by the studied mushroom species.

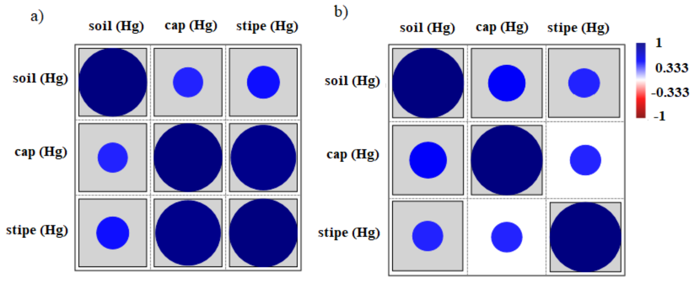

Figure 4 Spearman correlation matrices showing relationships between mercury concentrations in soil, caps, and stipes of (a) *Macrolepiota procera* and (b) *Cantharellus cibarius*. Circle size and colour intensity are proportional to the strength of the correlation coefficient (r).

The bioaccumulation factor (BAF) was used to determine the level of transition and accumulation of Hg from soil/substrate to the fruiting body of *M. procera* and *C. cibarius* (Figure 5). In the case of *M. procera*, all BAF values were higher than 1, for both the cap and the stipe. In the case of *C. cibarius*, BAF values were higher than 1 in only three cases, and only in the caps. Based on the results obtained, we can conclude that *M. procera* can be considered as mercury accumulator. This interpretation is further supported by the significant positive correlations observed between Hg concentrations in soil and mushroom tissues in both studied species. However, the stronger correlations and consistently higher BAF values observed for *M. procera* indicate a greater capacity for Hg uptake from the substrate. Mercury content in mushroom fruiting bodies is significantly influenced by the physicochemical properties of the substrate, particularly soil mercury concentration, pH, and organic matter content, which determine its mobility and bioavailability (**Demková *et al.*, 2021**; **Rzymski *et al.*, 2016**). Moreover, substantial interspecific differences in bioaccumulation capacity have been reported, with certain mushroom species exhibiting a higher tendency to accumulate mercury at elevated concentrations (**Melgar *et al.*, 2009**). Although individual species of fungi differ significantly in their ability to accumulate risk elements from the soil substrate, they are still considered exceptional bioaccumulators and indicators of soil quality (**Kumar *et al.*, 2024a**; **Ediriweera *et al.*, 2022**)

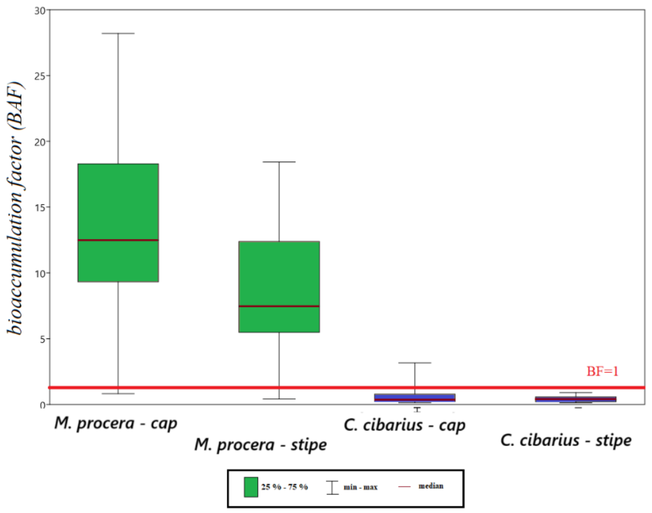

Figure 5 Bioaccumulation factor values determined in caps and stipes of two evaluated mushroom species.

The translocation factor (Qc/s) was used to express the mobility of mercury within mushroom fruiting bodies. The translocation factor values for *M. procera* ranged from 0.82 to 2.34 with an average value of 1.71, whereas the translocation in *C. cibarius* they ranged from 0.86 to 5.41 with an average value of 1.95 (Figure 6). Translocation factor values higher than 1 indicate the ability of the mushroom to translocate and potentially accumulate the element in the reproductive parts of the fruiting body (**Hussain *et al.*, 2022**). The average Qc/s values exceeding 1 in both species indicate preferential transport of Hg into caps. This finding is consistent with the significantly higher Hg concentrations observed in caps compared with stipes and suggests that consumption of caps may represent a greater source of dietary Hg exposure.Translocation occurs predominantly at the beginning of fruiting body formation (fructification), when metals are transported from the mycelium to the aboveground part (**Melgar *et al.*, 2009**). A high translocation factor poses an increased risk to consumers, as the caps, which people prefer to harvest and consume, can contain one-half to two-thirds more mercury than the whole fruiting body (**Árvay *et al.*, 2022**).

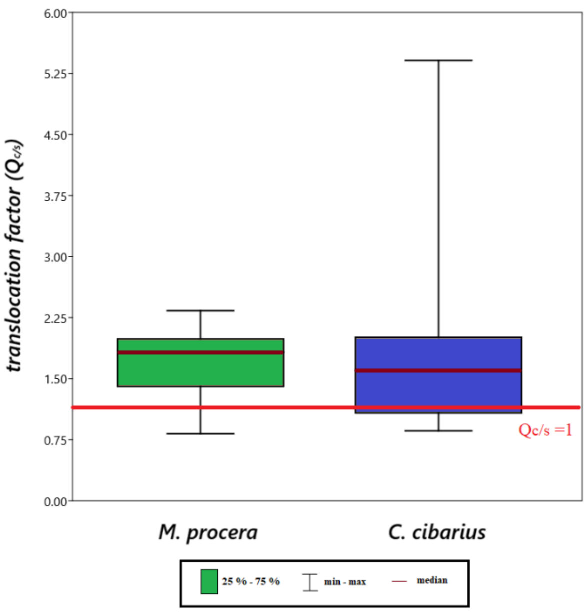

Figure 6 Translocation values determined in caps and stipes of two evaluated mushroom species.

The results obtained from the analysis of *M. procera* and *C. cibarius* showed that the % PTWI for caps reached substantially higher values compared to stipes (Figure 7). In *M. procera*, the % PTWI in caps ranged from 40.9 to 1314, while in stipes it ranged from 22.4 to 986. The 100% PTWI level was exceeded in 73.7% of cap samples and in 52.6% of stipe samples. In *C. cibarius*, the % PTWI in caps ranged from 1.10 to 125, and in stipes from 1.20 to 145. The 100% PTWI level in both caps and stipes of *C. cibarius* was exceeded at only one site, namely Smolnícka Huta. The highest values were determined in former mining areas, which are considered problematic in terms of elevated environmental contamination (**Pająk and Pietrzykowski, 2021**). As reported by several researchers the consumption of mushrooms collected near mining areas always poses increased health risks, (Árvay *et al.*, 2014; **Pastirčáková *et al.*, 2001**;). Target hazard quotient has been successfully used to evaluate whether intake of contaminants through food or other sources poses a potential health risk (**Gazzawi, 2025**). Target hazard quotient (THQ) values in caps and stipes of *M. procera* ranged between 0.78–25.1 and 0.42–18.9, respectively (Figure 8). In the case of *C. cibarius,* THQ values ranged from 0.02–2.38 and 0.02–2.77 for caps and stipes respectively. THQ makes it possible to quantify the risk associated with long-term exposure to contaminants that might otherwise manifest only after many years. It therefore allows for a better assessment of whether toxin concentrations are within acceptable limits (**Makroum *et al.*, 2026**). It should be noted that %PTWI and THQ values were calculated using average mercury concentrations and the average mushroom consumption rate reported for the Slovak population. Therefore, these indicators represent a standardized exposure scenario and should be interpreted as indicative estimates of potential health risk rather than precise population-level risk predictions.

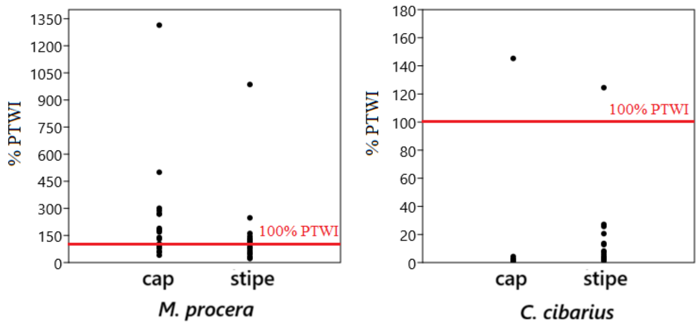

Figure 7 PTWI values determined in caps and stipes of two evaluated mushroom species.

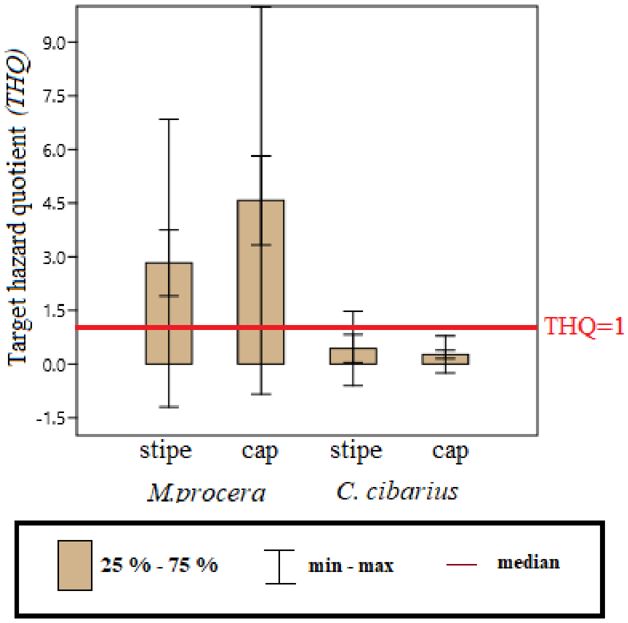

Figure 8 THQ values determined in caps and stipes of two evaluated mushroom species

CONCLUSION

The results of this study confirm that wild edible mushrooms can represent a significant pathway of mercury exposure, particularly when collected from environmentally burdened or historically contaminated areas. Elevated mercury concentrations were detected in several soil samples, especially in former mining regions, and these levels were directly reflected in the fruiting bodies of the analyzed mushroom species.

M. procera demonstrated a markedly higher capacity to accumulate mercury compared to *C. cibarius*, with bioaccumulation factor values consistently exceeding 1, classifying it as a mercury accumulator species. In contrast, *C. cibarius* showed considerably lower accumulation potential. In both species, mercury concentrations were significantly higher in caps than in stipes, indicating preferential translocation of mercury into the reproductive parts of the fruiting body, which are commonly consumed.

Health risk assessment revealed that in the case of *M. procera*, provisional tolerable weekly intake values were frequently exceeded, and target hazard quotient values above 1 indicated a potential risk of non-carcinogenic adverse health effects, particularly with long-term and regular consumption. The highest risk levels were associated with mushrooms collected from former mining sites.

Within the investigated Slovak localities, the findings highlight the importance of monitoring mercury contamination in edible mushrooms collected from historically contaminated areas and support the need for site-specific risk assessment. . Regular assessment of mercury levels in both soils and edible mushroom species is essential to ensure food safety and to minimize potential health risks for consumers.

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