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Contamination, bioconcentration and distribution of mercury in *Tricholoma* spp. mushrooms from southern and northern regions of Europe

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HIGHLIGHTS

- Mushrooms are a valuable food all over the world.
- Mercury concentrations in *Tricholoma* spp. from Croatia and Poland were determined.
- *Tricholoma* spp. mushrooms efficiently bio-accumulate mercury.
- Hg intake via *Tricholoma* spp. poses no health risk for studied regions.

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ABSTRACT

The contamination, bio-concentration and distribution of mercury (Hg) in wild mushrooms of the genus *Tricholoma* such as *T. equestre, T. portentosum, T. columbeta,* and *T. terreum* were studied, and the possible dietary intake and risk for human consumers in Europe was estimated. Mushrooms, together with the associated forest topsoils were collected from 10 unpolluted and geographically distant areas, far from local or regional emission sources, in Poland (2 sites) and Croatia (8 sites). The Hg contents were in the range 0.10 ± 0.06 to 0.71 ± 0.34 mg kg^{-1} dry matter in caps and 0.04 ± 0.02 to 0.38 ± 0.13 mg kg^{-1} in stems. The corresponding topsoil concentrations varied over a relatively narrow range between sites, from 0.013 ± 0.003 to 0.028 ± 0.006 mg kg^{-1} dry matter. Overall, the study results showed low levels of mercury both, in edible *Tricholoma* mushrooms and forest topsoils from background (unpolluted) forested areas in Croatia and Poland. The morphological distribution showed considerably greater concentrations of mercury in the caps relative to the stems with ratios ranging from 1.6 ± 0.6 to 3.9 ± 1.8. *T. equestre* showed good ability to bioconcentrate Hg, with bioconcentration factors (BCF) values in the range 18 ± 7 to 37 ± 18. The data suggests that *Tricholoma* mushrooms from unpolluted areas in southern and northern regions of Europe can be considered as a low risk food from the point of view of the tolerable Hg intake.

1. Introduction

Mercury (Hg) is a metallic element that occurs in the Earth's crust in trace concentration with an average concentration of 0.08 mg kg^{-1} in continental soils (Europe, Asia, Australia etc). In some globally diverse locations where concentrations in unpolluted soils rise to well above 0.1 mg kg^{-1}, anthropogenic activities contribute significantly to the geogenic background at a local/regional level, leading to large scale food and environment pollution with this element (Rytuba, 2003; Falandysz et al., 2015a, 2015b; Kojta et al., 2015; Falandysz, 2016, 2017; Beckers and Rinklebe, 2017; Lavoie et al., 2018). Mushrooms foraged from the wild are known to bioaccumulate Hg and can be locally and regionally, an important vector of Hg transmission to humans (Falandysz et al., 2019a).

The ongoing anthropogenic sources of mercury emission into

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the ambient environment are fossil fuels combustion, cement production, non-ferrous metal production, foundries and metal reclamation as well as its use in the chlor-alkali industry, batteries, fluorescent lighting, vinyl chloride monomer synthesis, recovery of gold by amalgamation in gold mining, dental amalgams, mining industry, former mercury—mining area etc. (Wilson et al., 2008; Árvay et al., 2014; Falandysz, 2016, 2017; UNEP, 2013). Mercury is considered to be a highly hazardous contaminant and unlike other vegetation in the terrestrial environment, it is efficiently absorbed from soil by mushrooms. Anthropogenic mercury emissions are considered to be important cause of Hg enrichment of forest soils during the last few decades (Demers et al., 2007; Falandysz et al., 2014a, 2014b). Awareness of Hg environmental toxicity has led to discontinuation of its use in many areas. However in other areas, the above mentioned anthropogenic sources continue unabated, resulting in the pollution of pristine forested lands (UNEP, 2013; Beckers and Rinklebe, 2017).

Mushrooms play a vital role in ecosystems because they are able to biodegrade plant and animal substrates on which they grow and they form an important part of the bio-geo-cycling of minerals in forest ecosystems (Falandysz and Borovička, 2013). The concentrations of metallic elements in soils that arise from anthropogenic pollution as well as the geogenic background and type of soil are important factors that influence elemental occurrence and contents in mushrooms. Other factors that have an influence include the type of soil, environmental and habitat related properties such as organic matter/carbon content of the soil and type of vegetation, and the individual characteristics of the fungi (species and its genetics, morphology, developmental stage, age of mycelium, production of ligands) (Falandysz, 2002; Sesli, 2006; Kojta et al., 2015; Nasr and Arp, 2011; Borovička et al., 2019; Falandysz et al., 2019b, 2019c; Kavčič et al., 2019). The biological determinants and molecular mechanisms of metallic and metalloid elements absorption and co-absorption by macrofungi are very well explained (Beneš et al., 2018; Borovička et al., 2019). In passive absorption, the mineral constituents can be found in cellular structures but active absorption results in more complicated assimilation (Melik, 2004). The low mass organic acids, e.g. citric and other acids, excreted by fungi can chelate poorly soluble mineral components of soil substrata to facilitate and accelerate their uptake by hyphae with resulting accumulation in the sporocarps (Falandysz and Borovička, 2013; Mleczek et al., 2016b).

Edible mushrooms are healthy foods that are highly useful for bowel function and also because of other nutritional properties including sensory features and organic and inorganic antioxidant constituents (including vitamin D and others), polysaccharides, and peptides (Cardwell et al., 2018; Kalač, 2019; Glamočija et al., 2015). Also, some fungal species are used in the prevention of diseases such as hypertension (Talpur et al., 2002) and hypercholesterolemia (Jeong et al., 2010) and to improve the immune system by the function of fungal polysaccharides (alfa-glucan, beta-glucan, beta-glucuronoglucan, manoksiloglucan, galaktosiloglucan etc) (Wasser, 2002). Additionally, they are relatively rich in nutritionally important bio-metals such as Zn, Cu and Mn and are good sources of selenium (Se) (Falandysz, 2013; Falandysz and Borovička, 2013). However, mushrooms also display a propensity to efficiently bioconcentrate both essential and certain toxic elements including radioactive species (Mleczek et al., 2016a; Szymańska et al., 2019). The edible tissues of certain mushrooms can be elevated in As, Ag, Cd, Hg, Pb or radiocaesium ($^{134/137}$Cs) depending on species and soil substrata condition (Byrne and Tušek-Žnidarič, 1990; Melgar et al., 2009; Petkovšek and Pokorny, 2013; Mleczek et al., 2015; Tucaković et al., 2018; Falandysz et al., 2019d, 2019e; Komorowicz et al., 2019) at concentrations that could be risky for human consumers. Generally, mushrooms are richer in minerals compared to leafy vegetables, pulses, grains and fruits (Turkdogan et al., 2003; Cocchi et al., 2006).

The accumulation of mercury in different macromycetes from a vast biodiversity of species of the fungal kingdom has been widely studied and measured in individual examples from Europe, China and Canada (Kojta et al., 2015; Kojta and Falandysz, 2016; Nasr et al., 2012). A number of studies focus primarily on Hg determination in mushrooms and the dose assessment from dietary intake (Falandysz et al., 2019a, 2019d). Generally, the wide range of variation in the Hg contents of foraged edible, medicinal and entheogenic mushrooms has been determined and it can often exceeded 1.0 mg kg^{-1} dry matter (dm) in unpolluted sites; indeed concentrations can reach up to 22 mg kg^{1} dm (Alonso et al., 2000; Demirbas, 2001; Tüzen and Soylak, 2005; Melgar et al., 2009; Jarzyńska and Falandysz, 2012; Maćkiewicz and Falandysz, 2012; Drewnowska et al., 2013; Falandysz, 2014; Falandysz and Drewnowska, 2015; Širić et al., 2016, 2017; Falandysz et al., 2015a, 2015b; 2016, 2018; Kojta et al., 2015; Saba et al., 2016a, 2016b; 2016c; Lipka et al., 2018). This can lead to contamination of prepared food, e.g. stir-fried meals made of bolete mushrooms collected across Yunnan in China has been found to contain Hg at elevated concentrations (Falandysz et al., 2019a, 2019d).

Other species that are efficient at sequestering mercury are *Amanita muscaria* (Demirbas, 2001; Drewnowska et al., 2013; Falandysz and Treu, 2019); *Boletus edulis* (Melgar et al., 2009; Falandysz et al., 2011; Širić et al., 2016); *Boletus reticulatus* (Širić et al., 2016, 2017) and some other *Boletus* species from China (Falandysz et al., 2019a, 2019d); *Imleria badia* (previous name *Xerocomus badius*) (Falandysz et al., 2012; Mleczek et al., 2015), *Macrolepiota procera* (Gucia et al., 2012; Širić et al., 2017); *Rugiboletus extremiorientalis* (previous name *Leccinum extremiorientalis*) (Falandysz et al., 2019a, 2019d); *Suillus gravillei* (Chudzyński et al., 2009), *Xerocomus* spp. (Falandysz et al., 2019a). Usually mercury content in mushrooms species of the genus *Tricoholma* varies between < 0.5 and 1 mg kg^{-1} (Tüzen et al., 1998; Demirbas, 2001; Melgar et al., 2009; Maćkiewicz and Falandysz, 2012; Falandysz et al., 2016; Širić et al., 2016, 2017), with the *Tricholoma portentosum* strain showing even higher levels of mercury bioaccumulation (Alonso et al., 2000).

Mercury is known as a serious carcinogenic metal toxin, which markedly weakens the function of the immune system and creates blockages in the autonomic nervous system. The characteristics of mercury poisoning vary depending on whether it is caused by elemental, inorganic or organic Hg, acute or chronic exposure to the poison, and the amount of Hg introduced (Mahajan, 2007; Silbernagel et al., 2011). Exposure to high concentrations of Hg can permanently damage the brain, kidneys and fetal development (ATSDR, 2013).

The *Tricholoma* mushrooms strains analyzed in this study were all edible types which can be prepared in various ways, - baking, frying, drying, pickling, souring, and freezing. This study investigates the occurrence and accumulation of Hg in *Tricholoma* mushrooms such as *T. portentosum, T. terreum, T. equestre* and *T. columbeta*. The Hg up-take potential and intake of toxicant from *Tricholoma* spp. consumption were also examined. Additionally reported literature data on Hg in *Tricholoma* spp. were also reviewed.

2. Materials and methods

2.1. Mushrooms and underlying forest topsoil

Certain mushrooms of the genus *Tricholoma*, e.g., *T. equestre*, are traditionally foraged and consumed, while some forms of this genus, which are relatively similar in appearance, are inedible or can be toxic (Rzymski and Klimaszyk, 2018). The 136 samples of four different species of mushrooms were collected from July to November, between years 2004 and 2014 in order to select better Hg accumulators. The samples were collected randomly, from eight large regions in Croatia and two in Poland. Samples ($n = 96$) of topsoil (0–10 cm) in which the mushrooms grew were also collected. The sampling locations were: Croatia – Trakošćan, Brezova gora (46°27′91.9″N 16°00′80.6″E); Medvednica (45°98′60.7″N 16°14′53.9″E); Maksimir (45°85′04.1″N 15°95′03.8″E); Dugi Dol, Karlovac (45°35′35.8″N 15°56′31.4″E); Ravna gora (45°33′52.8″N and 14°98′57.2″E); Skrad (45°42′59.2″N and 14°87′03.2″E); Island Krk (45°15′46.8″N and 14°56′13.3″E); Labinština (45°13′37.9″N and 14°12′75.1″E), and Poland - Biebrowo, Pobrzeże Słowińskie, Pomorskie Voivodeship, Gmina Kaliska. (Fig. 1)

The collected fruiting bodies of mushroom for this study were mature and in good body condition. Sampled mushrooms were freed from any visible plant and soil debris using a plastic disposable knife, during collection. In the laboratory, each specimens were separated into caps and stipes, siliced using a plastic knife and dried at 60 °C to constant mass with a commercial dryers that was commonly used for plant foodstuffs (MSG-01; MPM Product, Milanówek, Poland and Ultra FD1000 dehydrator, Ezidri, Australia) accordingly (Falandysz, 2014). Dried caps and stipes were ground with a laboratory mill Retch SM2000 to a powder and kept in polyethylene bags in dry condition until chemical analysis. Individual forest topsoil samples were pooled per site, air-dried at room temperature and sieved through a 2 mm pore size plastic sieve (Falandysz et al., 2003a). All analyzes were carried out in three replicates.

2.2. Analysis of mercury

Mercury concentrations in samples of mushrooms and forest soil were measured without acid digestion, using an atomic absorption spectrometer (AAS) mercury analyzer (AMA 254 Advanced Mercury Analyser, Leco, Poland). This analyzer uses direct combustion of the sample in an oxygen rich atmosphere. The measurement conditions used were: wavelength (253.65 nm); drying time (60 s); decay time (150 s); retention time (45 s); weight/volume of the sample (100 mg/100 mL); operating range (0.05–600 ng). Samples were analyzed in batches which also contained the calibration curve standards as well as two quality control samples that were over-spiked with known Hg concentrations. The detection limit (LOD) for Hg was 0.004 mg kg^{-1} dried product. Additionally, procedural blank samples, internal calibration and certified fungal reference materials were used with the aim of achieving high standards of analytical quality control and quality assurance (QC/QA) (Falandysz, 2017).

The bioconcentration factor (BCF) value was calculated as the quotient between the content of Hg in fungal material and the underlying topsoil layer. The results were evaluated using Statistica 10.0 software (Statsoft, USA). Descriptive data analysis included minimum value, maximum value, median, mean and standard deviation.

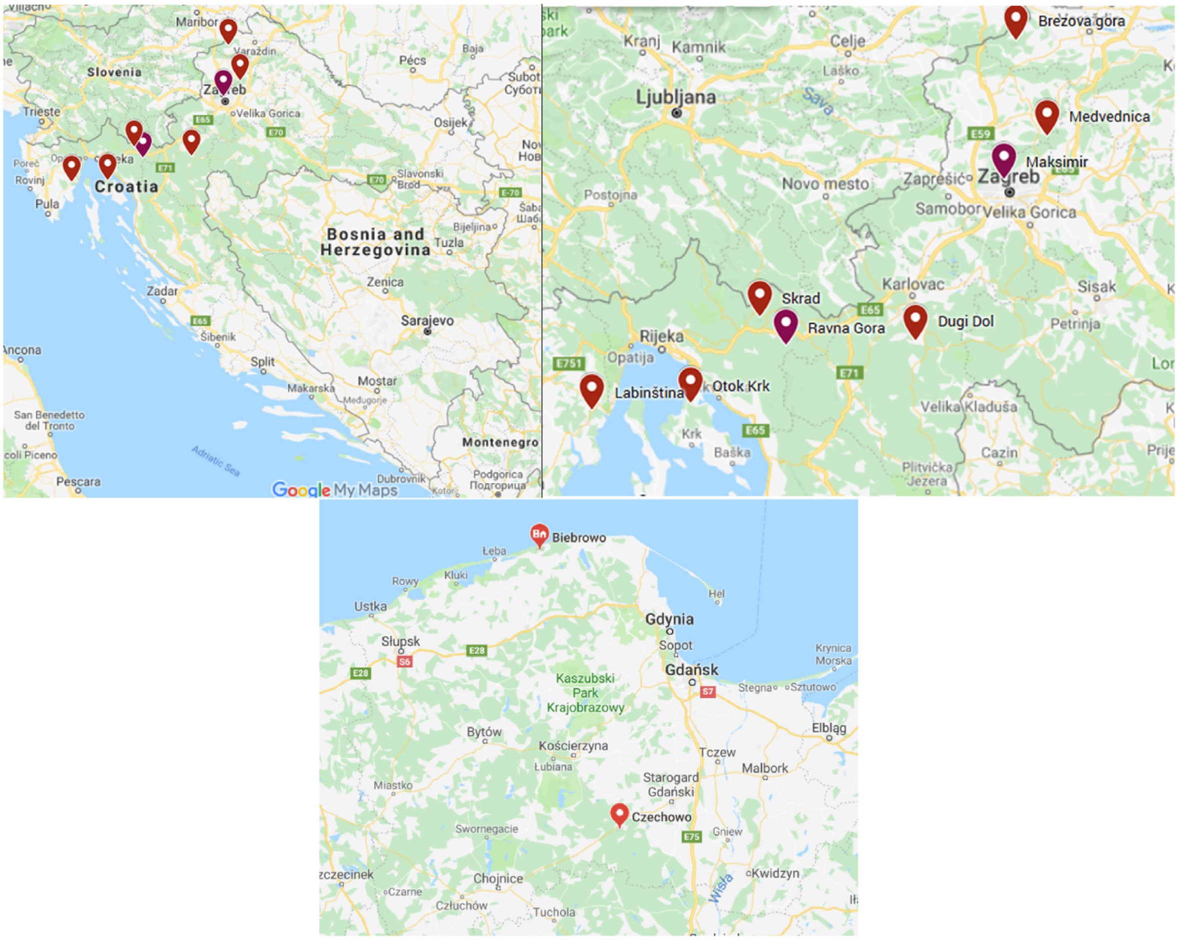

Fig. 1. Localities of the sampling species of mushrooms and associated forest topsoil.

3. Results and discussion

3.1. Mushrooms and topsoil

The *Tricholoma columbetta* (Fr.) P. Kumm, *Tricholoma equestre* (L.) P. Kumm., *Tricholoma potrentosum* (Fr.) Quel. and *Tricholoma terreum* (Schaeff.) P. Kumm. are ectomycorrhizal mushrooms which can be found in mixed forests. *Tricholoma portentosum* is most commonly associated with deciduous and coniferous trees. *Tricholoma terreum* is symbiotic with Scots pine *Pinus sylvestris* L. while *Tricholoma columbeta* often grows symbiotically with oak *Quercus* spp. and beech *Fagus sylvatica* L. *Tricholoma equestre* can be found below conifer trees, especially of the *Pinus* family.

Data on the concentrations of Hg (dry weight basis) in *Tricholoma* spp. and the associated top-soils are summarized in Table 1. Considerable differences were noted in the amounts of Hg accumulated by *Tricholoma* spp. from different locations but also associated to different topsoil concentrations (Table 1). There were low Hg concentrations in mushroom caps at ten locations, ranging between 0.10 ± 0.03 mg kg^{-1} dm in *T. terreum* (Skrad, Croatia) and 0.71 ± 0.34 mg kg^{-1} dm T. *equestre* (Biebrowo, Pobrzeże Słowińskie, Pomorskie Voivodeship, Poland), with wide variation from place to place. Mushrooms *T. portentosum* collected from Gmina Kaliska in Poland and *T. terreum* from Skrad in Croatia showed the same amount of Hg contamination, i.e., 0.10 ± 0.06 mg kg^{-1} dm and 0.10 ± 0.03 mg kg$^{-!}$, respectively.

Mercury in the forest soils was relatively low, ranging between 0.0094 and 0.046 mg kg^{-1} dm in Poland and 0.009–0.035 mg kg^{-1} dm in Croatia with mean values varying from 0.013 ± 0.003 mg kg^{-1} dm (Gmina Kaliska, Poland) to 0.028 ± 0.006 mg kg^{-1} dm (Labinština, Croatia). The Hg concentrations in the mushroom samples in this study are also generally low and to some extent, comparable to results in previous studies of Hg content in *Tricholoma* spp. and in substrates from other parts of Croatia (Širić et al., 2016, 2017) and Poland (Maćkiewicz and Falandysz, 2012). Interestingly, the mean Hg values were also comparable for soils determined in Croatia (Petrova gora) (0.021 ± 0.005 mg kg^{-1} dm) and the Biebrowo (0.022 ± 0.007 mg kg^{-1} dm) in Poland. These sampling sites have no known historical or current Hg emissions. According to Saba et al. (2016a), mercury accumulation in forest topsoil of Poland can be arise from anthropogenic sources (industrial processes, city sludge, treated seed, transport), and also natural sources of emissions (volcanic activity, volatilization from lithosphere).

Generally, levels of Hg in forest topsoil in Poland (0–15 cm) observed in this study are below 0.05 mg kg^{-1} dm (Falandysz, 2002, 2014; 2017; Falandysz et al., 2015a, 2015b; Saba et al., 2016a), while Širić et al. (2017) recorded 0.61 mg kg^{-1} dm levels of Hg of in Northwest Croatia. Similar, higher levels were reported in earlier research by Falandysz et al. (2012) from central, southern and south-western regions of Poland with topsoil Hg values > 0.05 mg kg^{-1} dw.

Bio-concentration factors (BCFs) were used to estimate any difference or similarity in the potential to accumulate metallic elements from forest topsoil by different mushrooms species. This data provides a general indication on whether a particular element is actively bio-concentrated (BCF > 1) or not (BCF < 1). The mean BCF values for caps were generally higher in relation to stipes. The median BCF value for the *T. equestre* in Biebrowo (Poland) was 30 with mean values 37 ± 18, which is about six times higher for the lowest established BCF values in *T. terreum*, Skrad (Croatia, median at 5.59). The BCF values showed that *T. portentosum* in Biebrowo, has a potential to accumulate mercury. However, the *Tricholoma* spp. if compared to certain other species of mushrooms showed weak potential to accumulate Hg. For example, very high bio-concentration factors were recorded for *Lycoperdon perlatum*, from 110 up to 420 (Falandysz et al., 2001, 2002), for *Boletus* spp. 126 to 491 (Melgar et al., 2009), *Cortinarius* spp. 22 to 150 (Falandysz, 2014) and *Macrolepiota* spp. up to 220 (Gucia et al., 2012; Kułdo et al., 2014).

T. equestre (common name - yellow knight) mushroom and soil samples that were obtained from a single site were more contaminated in contrast to *T. portentosum, T. terreum* and *T. columbetta* from other sites in Croatia and Poland (Table 1). As described in Table 2, fruiting bodies of *T. equestre* collected across Northern Poland showed a similar result to this study with a mean Hg values of 0.77 mg kg^{-1} dm.

Samples of *T. portentosum* were collected from four locations including one from Northern Poland and three from coastal Croatia. *Tricholoma portentosum* and topsoil from Gmina Kaliska localization in Poland showed lower contamination ($p < 0.05$) than the Croatian samples (Table 1). The Gmina Kaliska site (Poland), shows better bioavailability of Hg compared to other sampling sites in

Table 1
Sampling locations, presence of industry and distance from thermopower plants.

Sampling locations	Presence of industry	Distance from thermopower plants
Biebrowo, Pobrzeże Słowińskie, Pomorskie Voivodeship	Region of the landscape park; Baltic Coastal region, Agriculture and froestry to some degree, no industry, recreational/touristic region	Westerly winds dominate – no thermopower plants west and east of this point (nearest is about 150 km from the eastern side – in Gdańsk)
Gmina Kaliska, Pomorskie Voivodeship	Agriculture, forestry, no industry.	Westerly winds dominate – no thermopower plants west and east of this point (nearest is about 150 km from the eastern side – in Gdańsk)
Trakošćan, Brezova gora	Region on a board with Slovenia, recreational and touristic region, no industry.	No thermopower plants west and east of this point (nearest is about 120 km from the western side in Slovenia)
Medvednica	Region of the nature park; recreational and touristic region, no industry.	No thermopower plants west and east of this point (nearest is about 160 km from the western side in Slovenia)
Maksimir	Region of the nature park Medvednica; recreational and touristic region, no industry.	No thermopower plants west and east of this point (nearest is about 150 km from the western side in Slovenia)
Dugi dol	Agriculture and forestry area, no industry.	No thermopower plants west and east of this point (nearest is about 170 km from the western side in Slovenia)
Ravna gora	Forest area, a little agriculture, no industry.	No thermopower plants west and east of this point (nearest is about 120 km from the western side in Croatia, but was closed a long time ago)
Skrad	Forest area, a little agriculture, no industry.	No thermopower plants west and east of this point (nearest is about 130 km from the western side in Croatia, but was closed a long time ago)
Island Krk	Touristic regions, a little agriculture, no industry.	No thermopower plants west and east of this point (nearest is about 100 km from the western side in Croatia, but was closed a long time ago)
Labinština	Touristic regions, a little agriculture, no industry.	No thermopower plants west and east of this point (nearest is about 50 km from the eastern side in Croatia, but but was closed a long time ago)

Table 2

Mercury contents in *Tricholoma equestre, T. potrentosum, T. columbetta* and *T. terreum* and in related topsoils (mg kg^{-1} dry weight), Q$_{C/S}$ index and BCF values (mean ± SD, median and range).

Mushroom species, region, year and amount of specimens	Hg			BCFc	BCFs	$Q_{c/s}$
	Cap	Stipe	Soil			
Poland						
Tricholoma equestre Biebrowo, Pobrzeże Słowińskie, Pomorskie Voivodeship, 2004; $n = 15$	0.71 ± 0.34 0.60 (0.38–1.6)	0.38 ± 0.13 0.38 (0.19–0.61)	0.022 ± 0.007 0.020 (0.018–0.046)	37 ± 18 30 (10–76)	18 ± 7 19 (6.0–30)	2.0 ± 1.0 1.8 (1.0–5.6)
Tricholoma portentosum Gmina Kaliska, 2006; $n = 13(16)$	0.10 ± 0.06 0.085 (0.024–0.20)	0.055 ± 0.030 0.050 (0.016–0.13)	0.013 ± 0.003 0.013 (0.0094–0.018)	8.0 ± 4.3 9.1 (1.7–16)	4.6 ± 2.9 4.0 (1.1–11)	1.7 ± 0.4 1.7 (0.84–4.0)
Croatia						
Tricholoma portentosum Island Krk, 2012; $n = 10(12)$	0.23 ± 0.06 0.25 (0.13–0.29)	0.12 ± 0.019 0.12 (0.080–0.14)	0.016 ± 0.004 0.017 (0.011–0.023)	14 ± 3 15 (8.0–18)	6.9 ± 1.1 7.1 (4.9–8.6)	2.1 ± 0.7 2.1 (1.2–3.3)
Tricholoma portentosum Ravna Gora, 2012; $n = 9(11)$	0.34 ± 0.08 0.33 (0.23–0.48)	0.13 ± 0.03 0.14 (0.075–0.17)	0.018 ± 0.003 0.018 (0.012–0.026)	19 ± 5 18 (13–29)	7.3 ± 1.8 7.6 (4.2–9.4)	2.7 ± 0.8 2.6 (1.9–4.0)
Tricholoma portentosum Labinština, 2012; $n = 13(15)$	0.29 ± 0.14 0.23 (0.14–0.53)	0.18 ± 0.05 0.18 (0.12–0.26)	0.028 ± 0.006 0.029 (0.021–0.035)	10 ± 5 8.2 (5.1–19)	6.6 ± 1.7 6.3 (4.2–9.4)	1.6 ± 0.6 1.5 (0.79–2.6)
Tricholoma columbetta Trakošćan, Brezova Gora 2014; $n = 10(14)$	0.27 ± 0.13 0.28 (0.10–0.46)	0.12 ± 0.07 0.10 (0.02–0.23)	0.021 ± 0.005 0.022 (0.012–0,026)	13 ± 6 13 (4.9–22)	5.5 ± 3.2 4.8 (0.93–11)	3.2 ± 2.8 2.1 (1.1–11)
Tricholoma columbetta Medvednica, Stubaki 2014; $n = 14(19)$	0.32 ± 0.11 0.32 (0.15–0.47)	0.19 ± 0.06 0.18 (0.13–0.28)	0.024 ± 0.006 0.025 (0.011–0.031)	13 ± 5 13 (6.3–20)	8.0 ± 2.4 7.7 (5.2–12)	1.8 ± 0.8 1.4 (0.87–3.1)
Tricholoma terreum Maksimir, 2012; $n = 8(9)$	0.18 ± 0.05 0.18 (0.12–0.28)	0.09 ± 0.03 0.08 (0.06–0.13)	0.020 ± 0.006 0.019 (0.010–0.027)	9.1 ± 2.2 8.8 (6.1–14)	4.7 ± 1.5 4.1 (2.9–6.5)	2.2 ± 0.8 2.4 (0.98–3.1)
Tricholoma terreum Dugi Dol, Karlovac, 2012; $n = 9(12)$	0.12 ± 0.03 0.12 (0.08–0.18)	0.05 ± 0.02 0.05 (0.03–0.10)	0.015 ± 0.005 0.013 (0.009–0.022)	8.3 ± 2.1 8.0 (5.7–12)	3.6 ± 1.7 3.1 (1.7–6.8)	3.9 ± 1.8 2.6 (0.83–6.2)
Tricholoma terreum Skrad, 2013; $n = 10(13)$	0.10 ± 0.03 0.09 (0.07–0.16)	0.04 ± 0.02 0.03 (0.02–0.10)	0.017 ± 0.003 0.018 (0.012–0.021)	5.93 ± 1.71 5.59 (4.00–9.12)	2.61 ± 1.50 1.88 (1.29–5.74)	2.94 ± 1.64 2.85 (0.93–5.74)

Qc/s (cap to stipe Hg content quotient); BCFc and BCFs (bioconcentration factor values for caps and stipes, respectively).

Croatia, based on the BCF values for *T. portentosum* (Table 1). In Spain, in the Lugo region, Alonso et al. (2000) and Melgar et al. (2009) reported substantially higher Hg contamination in *T. portentosum.*

Samples of *T. columbetta* and *T. terreum* were collected from two and three sampling sites respectively, in Croatia. The fruiting bodies of these samples showed generally smaller Hg contents compared to results for the same species, as reported by Melgar et al. (2019), Širić et al. (2016) and Širić et al. (2017). On the other hand, *T. terreum* sampled from a site in Turkey showed mean Hg values of 0.217 mg kg^{-1} dm (Tüzen et al., 1998) which is similar to the results obtained in this study. However, in a later study, Demirbas (2001) reported extremely low concentrations of Hg in the *T. terreum* in Turkey (mean 0.06 mg kg^{-1} dm).

Morphologically, the considerably higher content of Hg in the mushroom caps compared to stipes that was determined in present study, is in good agreement with findings by other authors (Table 2). Other studies, e.g., Alonso et al. (2000) and Melgar et al. (2009), found substantial differences in the contents of Hg between the hymenophore and remaining part of the sporocarp in *T. portentosum, T. equestre* and *T. columbetta*. The mean concentrations of Hg in stipes were 2.4-fold lower than those in the caps (Table 1). A review of available results for Hg in species of the genus *Tricholoma* from 1995 to 2018 showed that the Hg contents were generally higher compared to the results of conducted research (Table 2).

3.2. Dietary intake

The concentration of mercury in different types of mushrooms can have a negative impact on consumers. The presence of toxic elements in wild edible mushrooms, amount of intake of these, nutritional convenience and potential risks are of direct relevance to consumers of this food source (Saba et al., 2016a). Mushroom species of genus *Tricholoma* can be prepared in numerous ways. According to Falandysz and Drewnowska (2015), boiling fresh mushrooms for a short time (15 min) does not substantially remove Hg form the flesh of fruiting bodies, or decrease the content of this element in mushroom meals relative to raw mushrooms (calculated on dry weight to dry weight data). Also, Wang et al. (2014) reported that varying the cooking procedures cause negligible loss of Hg from the meal. In estimating the potential risks from mercury intakes on consumers of caps of *Tricholoma* spp., JECFA has set a provisional tolerable weekly intake value - PTWI (0.004 mg kg^{-1} bw) for person of 70 kg body mass (JECFA, 2010). In this study, the median Hg concentrations in different mushroom caps were: *T. equestre* 0.60 mg kg^{-1} dm, *T. portentosum* 0.085–0.33 mg kg^{-1}, *T. columbetta* 0.28–0.32 mg kg^{-1}, and *T. terreum* 0.09–0.18 mg kg^{-1} dry weight (Table 1). These mercury concentrations, when expressed on a wet weight – fresh mushrooms (humidity content at 90%) vary from 0.0085 to 0.060 in Poland, and from 0.009 to 0.033 mg kg^{-1} in Croatia. The quantity of a single mushroom meal is estimated as 100–300 g and rarely higher than 500 g (Chudzyński et al., 2011). For fresh Tricholoma spp., the total intake by human consumption ranges between 0.1 kg and 0.5 kg (Table 3). The caps of *Tricholoma equestre* collected from Biebrowo in Poland forest show the highest Hg contamination levels. The intake estimated using median values of Hg in *Tricholoma* spp. foraged in Poland calculated for a person of 70 kg body weight, gave a range 0.0026–0.018 mg Hg potential dietary intake

Table 3
Mercury in *Tricholoma equestre, T. potrentosum, T. columbetta, T. terreum, T. matsutake and T. pessundatum,* and associated forest topsoil in Europe and Asia (China) (mean ± SD; mg kg^{-1} dry weight).

Mushroom species, region, year and amount of specimens	Hg			Reference
	Cap	Stipe	Soil	
Tricholoma equestre				
Poland, Pomerania, Hel Peninsula, Hel, 2004; $n = 15$	0.96 ± 0.32	0.62 ± 0.23	0.046 ± 0.007	A
Poland, Pomerania, Kolbudy Forest Inspectorate, Otomin, 2006; $n = 13(29)$[d]	0.77 ± 0.21	0.56 ± 0.26	0.019 ± 0.003	A
Poland, Pomerania, Rzecznica, 2003; $n = 10$	0.97 ± 0.10	0.69 ± 0.12	0.013 ± 0.002	A
Poland, Kociewie Land, Tucholskie forest, Łuby, 2001; $n = 14$	0.85 ± 0.06	0.65 ± 0.10	0.019 ± 0.003	A
Poland, Kujawy Land, Ciechocinek, 2004; $n = 15$	1.3 ± 0.7	1.1 ± 0.8	0.037 ± 0.002	A
Poland, Kujawsko-Pomorskie Voivodeship, Brzoza, 1999; $n = 14$	0.71 ± 0.11	0.68 ± 0.14	0.028 ± 0.006	A
Poland, Podlasie Land, Augustów, 1997/1998; $n = 16(80)$[d]	0.24 ± 0.08	0.17 ± 0.04	0.037 ± 0.009	AB
Poland, Podlasie Land, Augustów, 1998/1999; $n = 11$	0.25 ± 0.07	0.17 ± 0.04	0.036 ± 0.009	A
Poland, Podlasie Land, Augustów, 2006; $n = 15$	0.81 ± 0.10	0.63 ± 0.06	0.035 ± 0.001	A
Poland, Wdzydze Landscape Park, 1995–96; $n = 14$	0.12 ± 0.05	0.068 ± 0.030	0.020 ± 0.017	AC
Poland, Mazowsze Land, Kościelna Wieczfnia commune, 2006; $n = 13$	0.23 ± 0.06[a]		0.059 ± 0.028	A
Poland, Lubelskie Land, Chodelska Dale, Poniatowa, 1999–2001; $n = 13$	0.84 ± 0.42	0.49 ± 0.30	0.039 ± 0.007	A
Spain, Lugo, 2005/2006; $n = 6$	0.91 ± 0.54b	0.65 ± 0.39c	0.027	B
Tricholoma terreum				
Turkey, Sogultu	0.217[a]		WD	C
Turkey, Eastern Black Sea region	0.06 ± 0.02[a]		WD	D
Croatia, Medvednica, 2012; $n = 10$	0.34 ± 0.17	(0.44–0.36)	0.087 ± 0.012	E
Croatia, Trakošćan, 2012/2013; $n = 20$	0.49 ± 0.02	0.22 ± 0.03	0.061 ± 0.010	F
Poland, northeastern, Łukta and Morąg, 1997–1998; $n = 15(92)$ [d]	0.025 ± 0.034	0.014 ± 0.017	0.042 ± 0.016	G
Tricholoma columbetta				
Spain, Lugo, 2005/2006; $n = 7$	0.71 ± 0.28b	0.40 ± 0.04[c]	0.027	B
Tricholoma portentosum				
Spain, Lugo, 2005/2006; $n = 10$	1.1 ± 0.6[b]	0.65 ± 0.25[c]	0.027	B
Spain, Province of Lugo (1997); $n = 6$	1.31 ± 0.40	0.79 ± 0.36	WD	GA
Croatia, Medvednica, 2012; $n = 10$	0.93	0.66	0.087 ± 0.012	E
Croatia, Trakošćan, 2012/2013; $n = 20$	1.17 ± 0.06	0.82 ± 0.08	0.061 ± 0.010	F
Poland, Zaborski Landscape Park, 1997–1998; $n = 14$	0.18 ± 0.10	0.088 ± 0.052	0.025 ± 0.018	FA
Poland, Podlasie Land, Augustów, 1997/1998; $n = 16(80)$[d]	0.096 ± 0.030	0.030 ± 0.011	0.029 ± 0.007	AB
Poland, Wdzydze Landscape Park, 1995; $n = 14$	0.18 ± 0.07	0.082 ± 0.031	0.037 ± 0.005	AC
Poland, Wdzydze Landscape Park, 1996; $n = 3$	0.23 ± 0.02	0.12 ± 0.01	0.0034 ± 0001	AC
Sweden, Umeå, 1995; $n = 1(2)$	1.1	WD	0.027	FB
Tricholoma matsutake (S.Ito & Imai) Sing.				
China, Yimen in Yuxi prefecture, 2012; $n = 10$	0.73	0.44	WD	H
China, Yanshan, WenShan, 2012	1.0	0.62	0.27	H
Tricholoma pessundatum (S. Ito & Imai) Sing.				
China, Sichuan, Gongga mountain, 2012; $n = 24$	1.2	0.61	0.17	H
Tricholoma saponaceum (Fr.) P. Kumm				
Northeastern Poland, Łukta and Morąg, 1997–1998; $n = 15(87)$[d]	0.034 ± 0.020	0.015 ± 0.008	0.041 ± 0.016	G

WD – without data.
References (A, Maćkiewicz and Falandysz (2012); AB, Falandysz et al. (2002a); AC, Falandysz et al. (2003b); B, Melgar et al. (2009); C, Tüzen et al. (1998); D, Demirbas (2001); E, Širić et al. (2016); F, Širić et al. (2017); FA, Falandysz et al. (2002); FB, Falandysz et al. (2001); G, Falandysz et al. (2003); GA, Alonso et al. (2000); H, Falandysz et al. (2016)..

[a] Whole fruiting bodies.

[b] Hymenophore.

[c] Rest of fruiting body.

[d] Number of pooled individuals and total number of fruiting bodies (in parenthesis).

Table 4
Estimated consumption rate and probable dietary intake (PDI) for Hg from *Tricholoma* spp. mushroom species collected from Croatia and Poland by an individual of 70 kg body mass.

Estimated consumption (kg)	Area	PDI (mg *per capita*)	PDI (mg kg^{-1} body mass)
0.1	Croatia	0.0009–0.0033	0.000013–0.000047
0.1	Poland	0.00085–0.006	0.000012–0.000086
0.3	Croatia	0.0027–0.0099	0.000039–0.00014
0.3	Poland	0.0026–0.018	0.000036–0.0026

(PDI) or 0.000036–0.0026 mg Hg kg^{-1} body mass. For *Tricholoma* spp. collected in Croatia, the estimated Hg intake by a 70 kg person who consume 300 g caps, ranged from 0.0027 to 0.0099 mg (0.000039–0.00014 mg kg^{-1} body mass) (Table 3). However, the estimated weekly intake of total mercury from fish and shellfish from the Adriatic Sea was in the range 0.02–0.07 mg, representing 6–20% of the PTWI (Jureša and Blanuša, 2003). Thus, these intakes, estimated for *Tricholoma* spp. specimens collected from different areas in Croatia and Gmina Kaliska area in Poland were considerably below the provisional weekly tolerance limit (PTWI) of 0.004 mg kg^{-1} body mass. However, in the worst case scenario i.e. usigng the maximum values detected of Hg in caps of *Tricholoma equestre* collected from Biebrowo in Poland, intake can be closer to the PTWI 0.004 mg kg^{-1} body mass. Accordingly, in most cases the moderate consumption of analyzed genus *Tricholama* poses no risk to human health (Table 4).

4. Conclusions

The data on Hg contamination of mushrooms in Poland and Croatia shows clear differences in the ability of different species from different locations to accumulate this element. The mushroom *Tricholoma* spp. effectively absorbs mercury contained in forest topsoil in which its mycelium grows and develops. Comparison among two anatomical parts of fruiting bodies, i.e. the caps and stipes, in general showed the higher accumulation ability of the caps. A strong ability to bio-accumulate Hg, was clearly a feature all of the analyzed mushroom species (all BCF values $\gg 1$). The human dietary intake of Hg, estimated from the samples collected in this study of the genus *Tricholoma,* from localities in Croatia and Gmina Kaliska area in Poland, were considerably below the provisionally tolerable weekly intake. Moderate consumption of analyzed mushroom species can be considered safe from a toxicological point of view in most cases.

Disclaimer

The authors assert no conflict of interest.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

CRediT authorship contribution statement

Ivan Širić: Conceptualization, Resources, Methodology, Formal analysis, Data curation, Writing - original draft, Investigation, Writing - review & editing. **Jerzy Falandysz:** Conceptualization, Resources, Methodology, Funding acquisition, Formal analysis, Data curation, Writing - original draft, Writing - review & editing.

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