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Screening the Multi-Element Content of *Pleurotus* Mushroom Species Using inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES)

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Abstract Considering the worldwide popularity of cultivated *Pleurotus* mushrooms as food, analyses of their chemical composition are required to ensure product safety. The aim of this study was to compare the content of 62 elements in fruit bodies of six cultivated species of the genus *Pleurotus* (five strains of *Pleurotus ostreatus*, *Pleurotus eryngii*, *Pleurotus djamor*, *Pleurotus citrinopileatus*, *Pleurotus florida*, and *Pleurotus pulmonarius*), collected from their producers between 2009 and 2015. Only 31 elements (Al, As, B, Ca, Cd, Cr, Cu, Er, Fe, In, K, Lu, Mg, Mn, Na, Nd, P, Pb, Pt, Re, Rh, Sc, Se, Sr, Te, Th, Ti, Tm, U, Zn, and Zr) were detectable in the tested *Pleurotus* species in each year of their collection. The obtained results revealed three significantly diverse groups with similar abilities to accumulate 31 elements within 7 years of mushroom production. The species and strains were grouped as follows: *P. florida* and *P. pulmonarius* (first group); *P. ostreatus* HK35, *P. ostreatus* 930, *P. eryngii*, and *P. djamor* (second group); and *P. ostreatus* 80, *P. ostreatus* H195, *P. ostreatus* K22, and *P. citrinopileatus* (third group). In spite of differences between the tested *Pleurotus* species and strains, presented in the form of graphical Heatmaps, the intake of these mushrooms was not related with any health risk for consumers.

Keywords Edible mushrooms · *Pleurotus* · ICP-OES · Elemental analyses

Introduction

World production of mushrooms has increased nearly 25-fold over the last 35 years (1978–2012) and the global consumption of both wild and cultivated mushrooms has risen from approximately 1 kg in 1997 to 4 kg per capita in 2012 (Royse 2014). *Pleurotus* species, after *Agaricus* spp. are the second most cultivated and consumed mushrooms in the world. Between 1997 and 2010, *Pleurotus* spp. production increased from 876,000 to 6,288,000 t (data exclude production in India) (Chang 1999; Li 2012; USDA 2014; Sanchez and Mata 2012; Royse 2013; Yamanaka 2011). The greatest rise in production was reported in China and accounted for over 85 % of the world's total output in 2010. Approximately one fourth of China's mushroom production in 2010 was constituted by two species, *Pleurotus ostreatus* and *Pleurotus cornucopiae* (Li 2012). In Japan, production of *Pleurotus* spp. almost doubled between 1997 (13,300 t) and 2010 (39,600 t) while *Pleurotus eryngii* experienced the largest gains in production, increasing from 6734 t in 2000 to over 37,000 t in 2009 (Yamanaka 2011).

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Mushrooms from the genus *Pleurotus* (higher Basidiomycetes) are widely distributed around the globe with the exception of Antarctica (Zervakis et al. 2001). Under varying climate conditions different ecotypes of *Pleurotus* mushrooms have become established, among which commercially important ones include the following: *P. ostreatus* (oyster mushroom), *P. eryngii* (king oyster mushroom), *Pleurotus djamor* (pink oyster mushroom), *Pleurotus citrinopileatus* (golden oyster mushroom), *Pleurotus florida* (white oyster mushroom), and *Pleurotus pulmonarius* (Indian Oyster, Phoenix Oyster). However, phylogenetic studies have demonstrated that some of them, e.g., *P. ostreatus* and *P. florida*, represent in fact a single species (Gonzalez and Labarere 2000). The above-mentioned common names are used in trade. Altogether, *Pleurotus* spp. makes up almost 27 % of globally produced cultivated mushrooms (Chaubey et al. 2010; Royse 2014). Mushrooms of the genus *Pleurotus* break down cellulose, lignin, and hemicellulose during mycelium growth on materials of plant origin (Mikiashvili et al. 2004; Naik et al. 2012). Therefore, the most common cultivation substrates for this genus are easily accessible lignocellulose materials (Hassan et al. 2010; Moonmoon et al. 2010).

In Europe, in countries such as France, Italy, the Czech Republic, Slovakia, and Poland, cereal straw and agricultural and forestry wastes are used most often for cultivation of this genus (Akyüz and Yildiz 2008; Kirbag and Akyüz 2008). However, in countries with warmer climates such as Japan, Korea, China, or in South America, mostly rice straw, wood chips mixed with sliced straw, blends of various sawdusts with cotton husks, wheat bran, or soy in various proportions are employed (Qi et al. 2007; Akyüz and Yildiz 2008). The composition of the cultivation substrate greatly affects the growth of mycelium and yield, and to a large extent, determines the chemical composition of fruit bodies.

The fruit bodies of all *Pleurotus* species are generally characterized by a very good flavor and aroma and possess high nutritional value (particularly as regards to protein, fiber, potassium, calcium, magnesium, and iron content). Due to their low content of lipids and sugars, they are classified as a low-energy food item (Yujie et al. 2007; Bernaś et al. 2006; Kalač 2016). Moreover, they possess therapeutical potential owing to the biological activities of their metabolites (Daba et al. 2008; Dundar et al. 2008).

Mineral composition differs between species of *Pleurotus* mushrooms ranging from 4 to 10 % of dry matter (Deepalakshmi and Mirunalini 2014). The reported content of macro, micro, and trace elements of several *Pleurotus* species are presented in Table 1. The most commonly evaluated trace elements in *Pleurotus* mushrooms are Hg, As, Cd, and Pb. The standard element contents in various edible mushrooms from unpolluted areas, expressed as milligrams per kilogram in dry matter (dm), are as follows: Al 20–150, As <0.5–5, Ba 2–4, Cd 1–5, Co <0.5, Cr 0.5–5, Cu 20–100, Fe 50–300, Hg <0.5–5, Mn 10–60, Ni 15, Pb <5, Se <2, Sb <0.1, and Zn 25–200 (Kalač 2010). In general, all wood-decaying species of mushroom are low in Hg when compared to soil mushrooms. Mushrooms from the genus *Pleurotus* provide a good illustration of this. In the 1980s, Brunnert and Zadrazil (1983) found very high translocation rates of externally applied Cd and Hg in *P. djamor* and *P. ostreatus*. Recent research by Nnorom et al. (2012, 2013) has demonstrated that *P. ostreatus* contains between 0.028 and 0.031 mg kg^{-1} dm of Hg in caps and 0.028–0.0370 mg kg^{-1} dm in stipes, whereas *Pleurotus tuber-regium* contains from 0.024 to 0.048 mg kg^{-1} dm. The maximum content of Cd permitted by the European Union regulation in cultivated *Agaricus bisporus*, *P. ostreatus*, and *Lentinula edodes* is 2.0 mg kg^{-1} dm (assuming 90 % moisture) (Gucia et al. 2012). The reported Cd contents of *A. bisporus*, *P. ostreatus*, and *L. edodes* from Hungary showed the content of Cd in caps to be 0.17 ± 0.13, 0.91 ± 0.32, and 0.71 ± 0.48 mg kg^{-1} dm, respectively (Vetter et al. 2005); while on sale in Brazil, the content of Cd ranged between 0.011 and 0.23 mg kg^{-1} dm (Maihara et al. 2008). The maximum content of Pb permitted by the European Union regulation in cultivated *A. bisporus*, *P. ostreatus*, and *L. edodes* is 3.0 mg kg^{-1} dm (assuming 90 % moisture) (cited after Gucia et al. 2012). In cultivated species reported by Muñoz et al. (2005), contents of Pb were below 3.0 mg kg^{-1} dm (*A. bisporus* caps) 0.41 mg kg^{-1} dm, and *P. ostreatus* 0.91 mg kg^{-1} dm—data from Mexico). *Pleurotus sajor-caju* was reported to accumulate Pb in the amount of 4.24 mg kg^{-1} dm (Chauhan 2013). Studies of cultivated *P. ostreatus* from the European market by Costa-Silva et al. (2011) showed that Se was at 0.10 ± 0.01 to 0.26 ± 0.20 mg kg^{-1} dm, while the Se content of samples from the USA was 0.2 mg kg^{-1} dm (Hong et al. 2011). Cultivated mushrooms *A. bisporus*, *P. ostreatus*, *P. florida*, *P. eryngii*, and *L. edodes* contained As in amounts ranging between 0.009 and 0.210 mg kg^{-1} dm (Maihara et al. 2008). However, no maximum permitted levels have yet been set for inorganic As and methylmercury, although they are the most toxic species of As and Hg, respectively (Cordeiro et al. 2015). The research of Borovicka et al. (2011) showed that *P. pulmonarius* collected from unpolluted sites in the Czech Republic contained trace elements such as U and Th at levels of 3.25 and 11.4 µg kg^{-1} dm, respectively; Ag and Pb at 7.55 and 0.13 mg kg^{-1} dm, respectively. A study undertaken by Vetter (2005) showed Li content in *P. pulmonarius* to be 0.063–0.077 mg kg^{-1} dm. Available literature data on mineral elements in fruit bodies of *Pleurotus* species are collected in Table 1.

Table 1 Literature data on the selected elements content [mg kg^{-1} dry matter] in several *Pleurotus* species

Element	*P. ostreatus*[a]	*P. florida*[b]	*P. djamor*[c]	*P. citrinopileatus*[d]	*P. eryngii*[e]	*P. pulmonarius*[f]
Ca	190–1500	337–827	342	240	60–700	34–684
K	21,840–51,000	24,720	12,300–36,340	25,000	14,300–31,000	23,709–28,190
Mg	165–2300	134–359	316–1210	1600	1233–2000	183–2593
Na	250–1440	305	616	–	100–375	310–390
P	6180–13,390	6402	7432–7570	11,000	1300	5790–23,700
Ag	–	–	–	–	–	7.55
Al	14–85	–	–	–	–	–
As	0.025–1.0	0.73–0.83	–	–	0.009–0.35	–
Bi	1.9–9	–	–	–	–	–
Cd	0.28–5.39	0.22	0.028	–	0.011	–
Cr	0.1–16.3	–	1.63	–	1.3–22.6	–
Cu	19–50	10.6	14.5	16	6.83–48.5	–
Fe	33–550	62.7–432	14.0	95	24.2–190	75–154
Hg	0.5–2.0	–	–	–	–	–
Li	0.044–0.208	–	–	–	–	0.063–0.077
Mn	5–31.4	6.2–27	11.2	11	1.35–28.8	–
Ni	1.5–31.5	0.7	1.5	–	0.83	-
Pb	0.67–0.91	0.92	1.2	–	0.92–2.21	0.13
Se	0.11–0.55	0.132–0.48	1.1	–	–	–
Th	–	–	–	–	–	0.0114
U	–	–	–	–	–	0.00325
Zn	25–265	50.6–160	92.1	104	29.3–102	–

[a] *P. ostreatus* (Vetter 2005, Alam et al. 2008; Bernaś et al. 2006; Maihara et al. 2008; Muñoz et al. 2005; Kalač 2010; Costa-Silva et al. 2011; Zhu et al. 2011)

[b] *P. florida* (Alam et al. 2008; Mallikarjuna et al. 2013)

[c] *P. djamor* (Rodriguez Estrada and Royse 2007, Guo et al. 2007; Lee et al. 2009, Mallikarjuna et al. 2013)

[d] *P. citrinopileatus* (Rodrigues et al. 2015)

[e] *P. eryngii* (Rodriguez Estrada and Royse 2007, Akyüz and Kirbağ 2010; Zhu et al. 2011)

[f] *P. pulmonarius* (Oliveira Silva et al. 2002; Vetter 2003, 2005, Borovicka et al. 2011)

Recently, we have employed inductively coupled plasma optical emission spectrometer (ICP-OES) to investigate platinum group elements (Ir, Os, Pd, Pt, Rh, and Ru); light rare earth elements (Ce, Gd, La, Nd, Pr, and Sm); and heavy rare earth elements (Dy, Er, Eu, Ho, Lu, Tb, Tm, Sc, Y, and Yb) in wild mushroom species growing in Poland (Mleczek et al. 2016a). In the present, a broader, multi-element ICP-OES screening (total of 62 elements) of various commercially available *Pleurotus* mushrooms collected over a 7-year period (2009–2015) from Polish producers was conducted. Poland has a profound contribution to the world production of these mushrooms and this study is by far the most comprehensive insight into elemental composition of these foodstuffs.

Materials and Methods

Experimental Materials

The following 10 *P. ostreatus* species/strains were analyzed as follows: 1 = *P. ostreatus* 80, 2 = *P. ostreatus* H195, 3 = *P. ostreatus* HK35, 4 = *P. ostreatus* K22, 5 = *P. ostreatus* 930, 6 = *P. eryngii*, 7 = *P. djamor*, 8 = *P. citrinopileatus*, 9 = *P. florida*, and 10 = *P. pulmonarius*. The fruit bodies of *P. ostreatus* and *P. pulmonarius* strains were collected from 19 mushroom farms. The tested fruit bodies were obtained from each mushroom grower in several cultivation cycles each year in the period 2009–2015. The weight of a single fruit body sample amounted to 1 kg. Cultivation substrates of different compositions were applied by producers of oyster mushrooms in accordance with the cultivation method. The basic components of substrates were chopped wheat and rye straw. The substrates were supplemented with wheat bran, corncobs, sunflower seed hulls, and chalk by some mushroom growers depending on the cultivation method they used. The fruit bodies of *P. djamor*, *P. citrinopileatus*, *P. florida*, and *P. eryngii* were collected from 19 experimental cultivations in the Vegetable Crop Department of Poznań University of Life Sciences. As in the case of *P. ostreatus*, fruit bodies originated from several harvests during each year from the period 2009–2015. Each sample consisted of 0.25 kg of fruit bodies. *P. djamor*, *P. citrinopileatus*, and *P. florida* were cultivated on substrates of wheat straw with different additives. Supplements such as sawdust from deciduous trees, wheat and rye bran, flax shives, hemp shives, corn meal, soybean meal, and chalk were applied in different proportions in relation to the cultivation conditions. *P. eryngii*, on the other hand, was cultivated on substrates from a mixture of beech (*Fagus sylvatica* L.), alder (*Alnus glutinosa* Gaertn.), oak (*Quercus robur* L.). and birch (*Betula pendula* Roth) sawdust in different proportions. Sawdust substrates were supplemented as in the above-mentioned *Pleurotus* species. Gypsum was an additional component. Fruiting bodies were collected each year from several experimental cultivations in the amount of 0.25 kg in each replication.

Sample Processing

Ten samples of ten different *Pleurotus* species/strains were collected for analysis. Fruiting bodies from the first harvest of each year only were collected, as it was possible to collect the fruiting bodies of all 10 species/strains from the same places simultaneously (700 samples in total). In the case of the other harvests, fruiting bodies were not produced by the selected species so comparison of all 10 mushroom species was not possible and this material was not analyzed. Deionized ultrapure water (Milli-Q, Millipore, Saint Louis, USA) was used. Samples were dried at 105 ± 5 °C for 90 h in an electric oven (SLW 53 STD, Pol-Eko, Wodzisław Śląski, Poland) and ground in a laboratory Cutting Mill SM 200 (Retsch GmbH, Haan, Germany). Then, 0.400 ± 0.001 g of a dry sample was digested with concentrated nitric acid (65 % nitric acid, Merck, Darmstadt, Germany) in closed Teflon containers in the microwave digestion system Mars 5 Xpress (CEM, Matthews, USA). Samples were then filtered through paper filters and diluted with water to a final volume of 15.0 mL. Each of the samples was processed in 29 replicates.

Instruments and Quality Control

The inductively coupled plasma optical emission spectrometer Agilent 5100 ICP-OES (Agilent, USA) was used for the determination of 62 elements under common conditions, which are listed in the Supplementary data (Table S1). ICP commercial analytical standards (Romil, England) were applied for the calibration. The selected wavelengths and validation parameters were as follows: the detection limits obtained follow 3-sigma criteria on the level of 0.0X mg kg^{-1} dry matter (dm), precision and upper range of calibration curves are collected in Supplementary data (Table S2). Uncertainty for the complete analytical process (including sample preparation) was at the level of 20 %. Traceability was checked using the standard reference materials CRM S-1 = loess soil; CRM NCSDC (73349) = bush branches and leaves; CRM 2709 = soil; CRM 405 = estuarine sediments; and CRM 667 = estuarine sediments. The recovery (80–120 %) was acceptable for all the elements determined and detailed data is listed in Supplementary data (Table S3). Each of the samples was analyzed in triplicate.

Statistical Analysis

To compare the content of individual elements in the analyzed *Pleurotus* species within the tested period, the obtained data were illustrated using Heatmaps, where two-dimensional variables (mushroom species, element) were shown in color. Prepared figures allowed the similarities between tested mushroom species as regards to their ability to accumulate elements to be determined.

Cluster analysis allowed *Pleurotus* species to be separated as regards to element contents within the 7 years of studies (2009–2015) jointly but also separately for each year. This way to explain similarity inside groups was the most efficient among the groups. Using Ward Hierarchical Clustering and analysis of Euclidean distances, three plots were prepared.

Results

Among the 62 elements analyzed in the tested mushrooms, only 31 of them (Al, As, B, Ca, Cd, Cr, Cu, Er, Fe, In, K, Lu, Mg, Mn, Na, Nd, P, Pb, Pt, Re, Rh, Sc, Se, Sr, Te, Th, Ti, Tm, U, Zn, and Zr) were measurable in fruit bodies in each year of the collection period. In the case of the rest of the elements (Ag, Au, Ba, Be, Bi, Ce, Co, Dy, Eu, Ga, Gd, Ge, Hf, Ho, Ir, La, Li, Mo, Ni, Os, Pd, Pr, Rb, Ru, Sb, Sm, Tb, Tl, V, Y, and Yb) their content in numerous fruit bodies was below the limit of detection. The ability to accumulate the determined elements in the tested mushroom species varies widely (Table 2).

Table 2 Mean, the lowest and the highest content of analyzed elements [mg kg^{-1} dry matter] in *Pleurotus* species

Element	The lowest content		Mean	The highest content	
Al	*P. ostreatus* 930	0.60	4.39	44.14	*P. ostreatus* K22
As	*P. ostreatus* HK35	0.04	0.86	3.59	*P. eryngii*
B	*P. ostreatus* 80	0.07	3.65	24.80	*P. eryngii*
Ca	*P. florida*	37	157	2204	*P. eryngii*
Cd	*P. ostreatus* H195	0.06	0.47	5.54	*P. ostreatus* 80
Cr	*P. florida*	0.07	0.28	1.55	*P. ostreatus* K22
Cu	*P. eryngii*	3	15	54	*P. ostreatus* 930
Er	*P. pulmonarius*	0.06	0.38	1.60	*P. ostreatus* K22
Fe	*P. ostreatus* 930	18	105	775	*P. ostreatus* K22
In	*P. ostreatus* K22	0.05	0.31	1.07	*P. eryngii*
K	*P. florida*	10,970	26,050	56,710	*P. djamor*
Lu	*P. pulmonarius*	0.01	0.02	0.05	*P. ostreatus* 930
Mg	*P. pulmonarius*	918	2190	5145	*P. citrinopileatus*
Mn	*P. ostreatus* 930	4	12	27	*P. citrinopileatus*
Na	*P. citrinopileatus*	11	87	348	*P. eryngii*
Nd	*P. florida*	0.02	0.41	7.18	*P. eryngii*
P	*P. ostreatus* 930	4033	13,490	33,320	*P. citrinopileatus*
Pb	*P. ostreatus* H195	0.02	0.50	2.46	*P. ostreatus* 80
Pt	*P. ostreatus* 80	0.10	1.16	8.87	*P. ostreatus* K22
Re	*P. pulmonarius*	0.05	0.23	0.62	*P. citrinopileatus*
Rh	*P. pulmonarius*	0.01	0.10	0.60	*P. florida*
Sc	*P. pulmonarius*	0.01	0.03	0.13	*P. ostreatus* H195
Se	*P. ostreatus* 930	0.09	1.16	4.09	*P. citrinopileatus*
Sr	*P. pulmonarius*	0.02	0.37	6.63	*P. eryngii*
Te	*P. florida*	0.33	2.22	6.41	*P. ostreatus* HK35
Th	*P. eryngii*	0.01	0.15	0.38	*P. ostreatus* K22
Ti	*P. pulmonarius*	0.03	0.14	0.57	*P. ostreatus* K22
Tm	*P. pulmonarius*	0.01	0.05	0.19	*P. ostreatus* K22
U	*P. ostreatus* H195	0.06	1.00	4.09	*P. citrinopileatus*
Zn	*P. florida*	19	95	311	*P. djamor*
Zr	*P. pulmonarius*	0.01	0.04	0.54	*P. ostreatus* H195

The lowest contents of Al, Fe, Mn, P, and Se were observed in the strain *P. ostreatus* 930; those of Ca, Cr, K, Nd, Te, and Zn in *P. florida*; and of rarely reported elements Er, Rh, Sc, Tm and Zr in *P. pulmonarius*. *P. ostreatus* 930 displayed the highest content of Cu and Lu, and *P. florida of* Rh. A more distinct situation was observed for the other three *Pleurotus* species (*P. ostreatus* K22, *P. citrinopileatus*, and *P. eryngii*). The highest contents of Al, Cr, Er, Fe, Pt, Th, Ti, and Tm were determined in *P. ostreatus* K22; those of Mg, Mn, P, Re, Se and U in *P. citrinopileatus*, whereas *P. eryngii* was shown to contain the highest levels of As, B, Ca, In, Na, Nd and Sr. Additionally, these three species were characterized by the lowest contents of In (*P. ostreatus* K22), Na (*P. citrinopileatus*), and Th (*P. eryngii*).

Within the rest of the tested species/strains, *P. ostreatus* HK35 contained the lowest content of As and the highest Te level, *P. ostreatus* H195 the lowest content of Cd, Pb, and U and the highest Sc level. It is worth emphasizing that the highest content of Cd and Pb was observed in fruit bodies of *P. ostreatus* 80. *P. djamor* was the most effective accumulator of K and Zn.

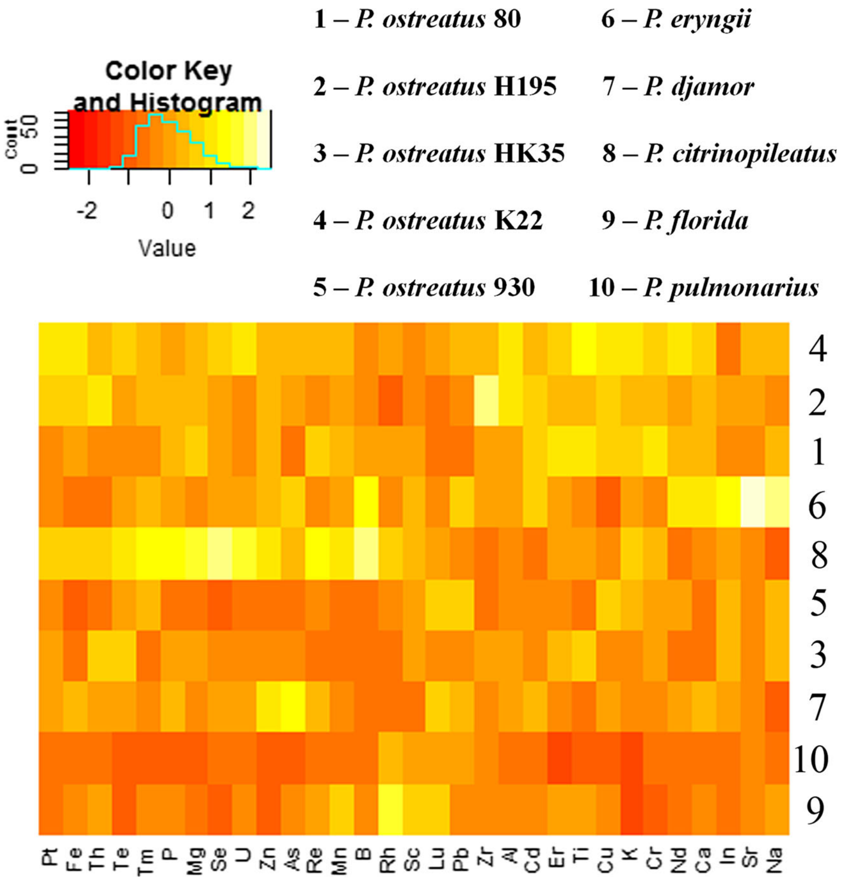

Fig. 1 Comparison of *Pleurotus* species with regard to mean content of 31 elements (Heatmap) within 2009–2015

To show the real similarities and differences in the content of particular elements determined in fruit bodies throughout the years of sample collection, a Heatmap was performed and the obtained results are presented in Fig. 1.

The same color in column indicates no differences in the content of the same element between tested *Pleurotus* species. This presentation was widened to show how these similarities or differences were revealed in particular years of mushroom production (Supplementary data, Fig. S1). To illustrate general differences between these species as regards to their content of all the analyzed elements, cluster analysis was used for preparation of the cluster dendrogram presented in Fig. 2.

Three significantly diverse groups of *Pleurotus* species were assigned. *P. florida* and *P. pulmonarius* were included in the first group as significantly different from the rest. The second group comprised *P. ostreatus* HK35, *P. ostreatus* 930, *P. eryngii*, and *P. djamor* while the third included *P. ostreatus* 80, *P. ostreatus* H195, *P. ostreatus* K22, and *P. citrinopileatus*. The differences between the tested mushroom species/strains and their insertion into the uniform groups in particular years are presented in Fig. 3.

The significance of the presented data relates to the variability in mushroom species assigned to the groups in particular years. The clearest relationships were observed for two

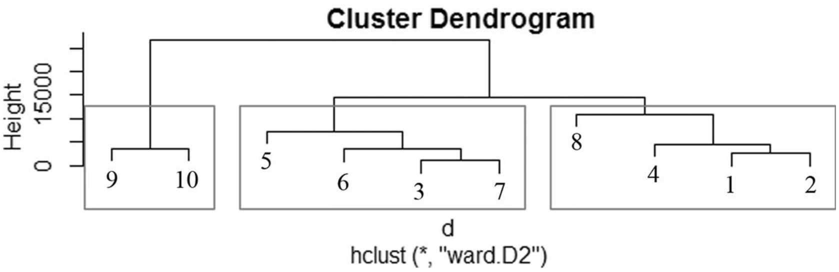

Fig. 2 A hierarchical tree plot showing the groups of mushrooms characterized by a high similarity in the accumulation of all 31 elements jointly. *1 P. ostreatus* 80; *2 P. ostreatus* H195; *3 P. ostreatus* HK35; *4 P. ostreatus* K22; *5 P. ostreatus* 930; *6 P. eryngii*; *7 P. djamor*; *8 P. citrinopileatus*; *9 P. florida*; *10 P. pulmonarius*

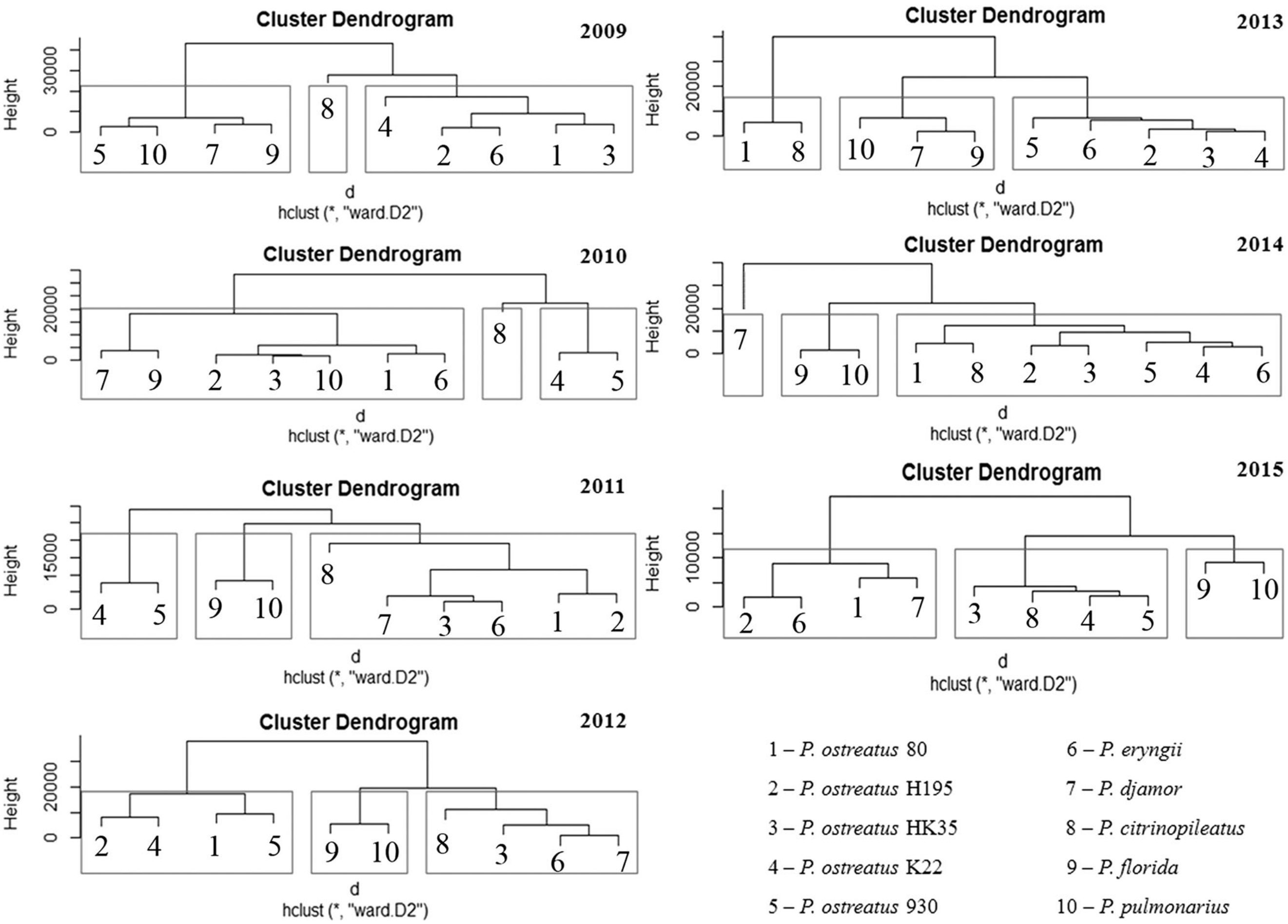

Fig. 3 A hierarchical tree plot showing the groups of mushrooms characterized by a high similarity in the accumulation of all 31 elements jointly in particular years of mushroom cultivation

pairs of mushrooms: *P. ostreatus* 80 and *P. ostreatus* H195 and *P. florida* and *P. pulmonarius*. In the former pair, the content of all 31 elements in *P. ostreatus* 80 and *P. ostreatus* H195 fruit bodies collected in all years was similar with the exception of 2013, while the latter pair of *P. ostreatus* strains belonged to the same group every year. These data suggest that the substrate used by the mushroom producers for the cultivation of *Pleurotus* mushrooms plays a considerable role.

Discussion

Content of Elements in Pleurotus Fruit Bodies

The present study is so far the broadest investigation of the multi-element content in fruit bodies of cultivated mushrooms belonging to the *Pleurotus* genus. As demonstrated, the investigated species/strains generally varied from each other in terms of element accumulation, not only in observed contents but also in their rank of order. Furthermore, the same species/strains collected during different seasons were also found to vary mutually. Hence, the present study suggests that the commercial substrates available for mushroom producers can exert a considerable influence on the chemical composition of *Pleurotus* mushroom fruit bodies. In fact, the number of different materials and their additions that can be used to cultivate these species is very large. The composition of substrate plays a crucial role in mineral element uptake and accumulation, including toxic elements, in consumable mushroom parts. As previously shown, the addition of various elements, both non-essential and nutritional, to cultivation substrate usually results in a proportional increase in their content in fruit bodies (Mleczek et al. 2016b; Rzymski et al. 2016a, b). Considering the popularity of *Pleurotus* mushrooms for global mushroom production as a foodstuff, it is of high interest to screen their chemical content and to assess whether they may contain potentially toxic levels of certain elements.

The mean contents of detected elements in the fruit bodies of the investigated species generally decreased in the following order K > P > Mg > Ca > Fe > Na > Zn > Cu > Al > Mn > B > Pt > Nd > Sr > Te > Cd > Se > U > As > Pb > Er > Cr > In > Re > Rh > Ti > Zr > Th > Tm > Sc > Lu. Based on the identified contents, five main groups of elements could be classified (expressed per dry weight): (i) those exceeding 1000 mg kg^{-1} (K, P, Mg, and Ca); (ii) ranging from 100 to 1000 mg kg^{-1} (Fe, Na, Zn); (iii) ranging from 10 to 100 mg kg^{-1} (Cu, Al, Mn, B); (iv) ranging from 1 to 10 mg kg^{-1} (Pt, Nd, Sr, Te, Cd, Se, U, As, Pb, Er, Cr, In); and (v) below 1 mg kg^{-1} (Re, Rh, Ti, Zr, Th, Tm, Sc, Lu).

Characteristics of Potential Health Risks

Apart from rate of element accumulation in fruit bodies of the investigated species, it is of high interest to assess whether the identified levels of toxic or potentially toxic elements can pose a potential risk for human health following consumption of such mushrooms. Besides nutritionally essential elements, the present study evaluated the content of Al, As, Cd, and Pb, which can be considered as non-specific for human metabolism and further, toxic at certain levels. Moreover, rare earth elements (REEs), whose health risk is still unknown, require an evaluation. REEs are increasingly emitted mostly from industry and include such elements as La, Ce, Pr, Nd, Sm, Eu, Gd, Tb, Dy, Ho, Er, Tm, Yb, Lu, Sc, and Y (Gambogi 2011; Li et al. 2013). The present study evaluated the content of five REEs (Er, Nd, Tm, Lu, and Sc). According to our knowledge, the only regulation for total REEs in foodstuffs was set in China at 0.7 mg kg^{-1} fresh weight (SAC 2005, 2012). The total mean content of five REEs in the investigated *Pleurotus* mushrooms exceeded this level (0.9 mg kg^{-1} dry weight). Most striking, however, was the relatively high content of Nd, which at its maximal level found in *P. eryngii*, was over 8 mg kg^{-1} dry weight. This is over three times more than the highest Nd level determined so far in wild mushrooms (Mleczek et al. 2016a). This finding indicates that even some cultivated mushrooms may be a significant source of Nd—an element which can act as an anticoagulant and which is considered to be moderately toxic, even though further evaluation of its biological activity is necessary.

The provisional tolerable weekly intake (PTWI) for Al is 2 mg kg^{-1} bodyweight (JECFA 2012). Considering an average single serving is 300 g of fresh mushrooms, i.e., about 30 g of dry matter (as a 10 % level is used for calculations with an unknown factual dm level), the mean and maximal content determined in the investigated mushrooms (4.4 and 44.1 mg kg^{-1} dm) would contribute insignificantly to PTWI for an adult weighing 60 kg.

The content of As was also too low to contribute significantly to As exposure through consumption of the investigated species. This is important if one considers that *P. eryngii* and *P. ostreatus* were previously shown to accumulate very high levels of As from artificially contaminated substrate (Mleczek et al. 2016b). The PTWI of total As was set at 15 μg kg^{-1} bodyweight (JECFA 1988) but this was later revised because the lower limit on the benchmark dose for a 0.5 % increasing incidence of lung cancer was in a similar range (FAO and WHO 2011). Nevertheless, the maximum level of As in *Pleurotus* mushrooms would account for 11.9 % of PTWI (considering that 30 g dm would be consumed by a 60-kg adult). It is also important to highlight that the toxicity of As largely depends on its chemical species; therefore, the risk of any adverse effects from As, following the consumption of the investigated mushrooms, was very low.

The provisional tolerable monthly intake (PTMI) of Cd was set at 0.025 mg kg^{-1} bodyweight (JECFA 2011), which accounts to 1.5 mg per month for a 60-kg adult. In this case, the risk of adverse Cd exposure was also very low if one considers that the mean and maximum level of this element found in the investigated mushrooms would account for 0.7 and 11.3 %, respectively (when 30 g dm is consumed by a 60-kg individual).

It should, however, be pointed out that Pb contents in fruit bodies of the investigated mushrooms were relatively high. The WHO determines the PTWI of lead at 0.025 mg kg^{-1} bodyweight (and equivalent of 0.0036 mg kg^{-1} body weight per day) (JECFA 2011), i.e., 1.5 mg weekly (and 0.21 mg daily) for an adult weighing 60 kg. The maximum determined Pb content of 2.46 mg kg^{-1} dm in *P. ostreatus* 80 would thus deliver a dose of 0.074 mg from a serving of 300 g of the fresh fruit bodies. This indicates that some substrates used for *P. ostreatus* cultivation could be contaminated with Pb. It is worth noting that the screening of commercially available mushrooms in China also revealed that *P. ostreatus* contained the highest Pb levels—a finding related to the substrate applied for cultivation (Huang et al. 2015). Overall, this highlights the need to check Pb content in these mushrooms and more particularly, the composition of substrates used for their cultivation. On the other hand, one should consider that the bioaccessibility of toxic elements from the investigated mushrooms in the human gastrointestinal tract can be reduced by cooking treatments such as boiling or microwaving with water (Sun et al. 2012).

Conclusions

The present study indicated that there are significant differences in element accumulation by different commercially cultivated species of the genus *Pleurotus*. Importantly, the levels of most of toxic elements were low apart from Pb contents, which were of some concern and highlighted the need to check substrates used for cultivation of these mushrooms. The strikingly high content of Nd, an element of as yet incompletely elucidated toxicity, was found in some mushrooms.

Compliance with Ethical Standards

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Conflict of Interest Marek Siwulski declares that he has no conflict of interest. Mirosław Mleczek declares that he has no conflict of interest, Piotr Rzymski declares that he has no conflict of interest, Anna Budka declares that he has no conflict of interest, Agnieszka Jasińska declares that he has no conflict of interest, Przemysław Niedzielski declares that he has no conflict of interest, Pavel Kalač declares that he has no conflict of interest, Monika Gąsecka declares that he has no conflict of interest, Sylwia Budzyńska declares that he has no conflict of interest, Patrycja Mikołajczak declares that he has no conflict of interest.

Ethical Approval This article does not contain any studies with human participants or animals performed by any of the authors.

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