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Research Article ACCUMULATION OF METALS IN SOME WILD AND CULTIVATED MUSHROOM SPECIES

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ABSTRACT

In this study; the contents of some trace elements (Mg, Al, Ca, Mn, Fe, Co, Ni, Cu, Zn, Se), toxic heavy metal contents (Cr, As, Cd) and lead isotopes (206Pb, 207Pb and 208Pb) were determined in some wild edible (Amanita caesarea (Scop.) Pers., Cantharellus cibarius Fr., Craterellus cornucopioides (L.) Pers., Fistulina hepatica (Schaeff.) With., Meripilus giganteus (Pers.) P. Karst) and cultivated mushroom (Agaricus bisporus (J.E. Lange) Imbach, Pleurotus ostreatus (Jacq.) P. Kumm.) species. The trace elements and toxic heavy metal contents were found lower than the upper limits in both wild edible mushrooms and cultivation mushrooms. The elemental composition of the wild mushrooms and the cultivated mushrooms obtained from different firms was found to be statistically different (p<0.05) from each other. C. cornucopioides was revealed with the highest Ca, Mn, Ni, Cu, Zn, ²⁰⁸Pb and Cr contents. The metal accumulation in wild mushrooms was higher than the cultivated mushrooms except Al and Se. All investigated parameters revealed that there was not any health risk associated with the consumption of the analyzed wild and cultivated mushrooms.

Keywords: Cultivated mushroom, wild mushroom, heavy metal, trace element, lead isotopes.

1. INTRODUCTION

People have used mushrooms as a nutrient for centuries thanks to their flavor, specific aroma, chemical composition and nutritional value [1]. Some mushroom species also have medicinal properties such as antioxidant [2] antimicrobial [3, 4], antifungal and anticancer [4, 5]. Due to the more reliable source of supply, cultivated mushrooms are generally preferred to wild mushrooms. Among over 20 cultivated species, the most cultivated mushroom type is A. bisporus (champignon), followed by Lentinula edodes (Berk.) Pegler (shiitake) and P. ostreatus (ovster mushroom) [6] On the other hand, wild-growing mushrooms have been a delicacy in many countries, for a long time. Given the climatic conditions, Turkey has a great potential for wild edible mushroom species and has become an important exporter.

On the other hand, the metal pollution accumulated on foods has become a serious problem, nowadays [7]. Many elements, such as cadmium, mercury, arsenic, etc., are toxic to living organisms especially at high concentrations [8]. Mushrooms have the ability to accumulate both the natural and anthropogenic radionuclide [9, 10] and heavy metals [11]. Therefore, determining

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the metal content in mushroom is very important to know the risk level that affects the human health directly or ind irectly. It was reported that when using industrial wastes/sludge's for mushroom cultivation, careful attention must be paid to the translocation of toxic substances from substrates to mushrooms [12].

Undoubtedly, several studies about heavy metal content of mushrooms have been published [8-13]. However, there are a few studies comparing the metal contents of wild and cultivated mushrooms which cultivated different mediums. For that purpose; the metal contents of *A. bisporus* and *P. ostreatus* cultivation mushroom species obtained from different commercial companies in the Trabzon province (Turkey) were determined and compared with the metal contents of wild mushrooms (*A. caeserea, C. cibarius, C. cornucopioides, F. hepatica, M. giganteus*) in the Kastamonu province (Turkey).

2. MATERIALS AND METHODS

The species of mushrooms, their habitats, locations and growing forms are given in Table 1.

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No	Mushroom species	Habitat and Location	Edibility	Growing Form
1	A. bisporus (A)	Substrate, Trabzon	Edible	Cultivated
2	A. bisporus (B)	Substrate, Trabzon	Edible	Cultivated
3	A. bisporus (C)	Substrate, Trabzon	Edible	Cultivated
4	A. bisporus (D)	Substrate, Trabzon	Edible	Cultivated
5	P. ostreatus	Substrate, Trabzon	Edible	Cultivated
6	A. caesarea	Soil, Kastamonu	Edible	Wild
7	C. cibarius	Soil, Kastamonu	Edible	Wild
8	C. cornucopioides	On soil, Kastamonu	Edible	Wild
9	F. hepatica	On tree trunk, Kastamonu	Edible*	Wild
10	M. giganteus	On tree trunk, Kastamonu	Edible	Wild

Table 1. The species of mushrooms, their habitats, locations and growing forms

*It is known as poisonous in the current location

Wild mushroom specimens were collected from the Doğanyurt district of Kastamonu province (42° 00' 00" N, 33° 27' 01" E). The ecological and morphological characteristics were noted, and the specimens were photographed in their natural habitat. Later, mushrooms were identified based on their morphological characteristics and then dried for future uses. In the study, two different cultivated mushroom species (*P. ostreatus, A. bisporus*) were also investigated. *P. ostreatus* mushroom was obtained from one (1) commercial firm and *A. bisporus* mushroom was obtained from four (4) different commercial firms in Trabzon/Turkey. Ethically, all commercial mushroom firms were coded as A, B, C and D.

Samples preparation

All the mushroom samples were sliced and dried in a drying mechanism until they were completely dehydrated. The samples were crushed to pass through a 40 mesh sieve.

Samples treatment

Mushroom samples (0.5 g) were digested in a mixture of 5 mL of HNO_3 (65%), 2 mL of HCl (37%) in a microwave digestion system for 31 min and diluted to a 50 mL volume with deionized water and then samples were filtered through 0.45 micron filters These samples were analyzed by inductively coupled plasma mas s spectrometry (A Bruker 820-MS ICP-MS).Details regarding the instrumental and operating conditions are described in Table 2.

Table 2. Instrumental and operating conditions								
Analysis Modes		Ion Optics (volt)						
Analysis Type	Quantitative	Version Number	7					
Acquisition Mode	Steady State	First Extraction Lens	-1.00					
Scan Mode	Peak Hopping	Second Extraction Lens	-176.00					
Spacing	Coarse	Third Extraction Lens	-247.00					
Points/Peak	1	Left Mirror Lens	49.00					
Scans/Replicate	10	Right Mirror Lens	28.00					
Replicates/Sample	10	Bottom Mirror Lens	33.00					
Plasma		Corner Lens	-190.00					
Plasma flow	18.00 L/min	Entrance Lens	-1.00					
Auxiliary flow	1.80 L/min	Fringe Bias	-2.50					
Sheath Gas Flow	0.17 L/min	Entrance Plate	-34.00					
Nebulizer flow	0.92 L/min	Detector Focus	True					
Sampling depth	6.50 mm	Pole Bias	0.00					
Power	1.40 kW	CRI (mL/min)						
Pump rate	5 rpm	Skimmer Cone	Off					
Stabilization delay	10 sec	CRI Skimmer Gas Flow	0 mL/min					
		Sampler Cone	Off					
		CRI Sampler Gas Flow	0 mL/min					
Sampling								
Aerosol generation	Nebulizer	Spray Chamber Cooling	On					
Source	Auto sampler	Spray Chamber Temp.	3.00 °C					
Fast pump during sample delay/rinse	On	Sample uptake delay	40 sec					
Enable device control	Off	Smart Rinse	No					
Probe height	0 mm	Switch Delay	Off					
Premix	Off	Scan time	841 msec					
Rinse time	10 sec	Replicate time	8.41 sec					

Table 2. Instrumental and operating conditions

Statistical analysis

The data were recorded as means \pm standard deviations of ten replicates and analyzed by using Statistical Package for Social Sciences (SPSS version 23.0). The data were analyzed with ANOVA and tests of significance were carried out using Duncan's multiple range tests. Pearson correlation coefficient was used to determine the relationship between the metals.

3. RESULTS AND DISCUSSIONS

The trace elements, toxic heavy metal and lead isotopes contents of mushrooms are presented in Table 3 and Table 4, respectively. Mg is an important element because many enzymes need it as co-factor [14]. The highest Mg content (176.1 mg/kg) was found in *M. giganteus* mushroom. Mg contents of wild and cultivated mushrooms are not significantly different from each other according to the Duncan's multiple range test.

Mushroom	Mg	Al	Ca	Mn	Fe	Co	Ni	Cu	Zn	Se
A. bisporus	123.5	28.9	286.1	12.2	245.3	0.19	14.6	22.6	39.8	4.54
(A)	(8.4)a	(1.4)e	(19.2)b	(0.73)g	(14.6)c	(0.006) bc	(1.1)bc	(1.2)b	(2.1)a	(0.17)d
A. bisporus	121.7	14.7	641.8	9.9	153.1	0.17	13.7	39.8	61.3	2.56
(B)	(5.4)a	(0.7)ab	(43.3)cd	(0.72)f	(3.3)ab	(0.015) bc	(0.9)b	(2.8)g	(4.8)d	(0.22)a
A. bisporus	115.1	78.8	842.0	7.6	104.5	0.16	11.6	36.7	53.3	2.98
(C)	(3.2)a	(8.6)g	(77.8)f	(0.92)cd	(12.7)a	(0.009) bc	(0.9)ab	(2.7)fg	(3.4)cd	(0.14)bc
A. bisporus	118.4	16.7	837.5	9.1	137.4	0.11	6.26	28.6	43.6	3.08
(D)	(4.1)a	(1.7)ab	(54.0)f	(0.27)ef	(6.9)ab	(0.004) ab	(0.7)a	(2.3)cd	(2.8)ab	(0.12)bc
P. ostreatus	169.1	24.6	140.4	8.6	169.1	0.17	15.3	15.0	73.3	3.07
	(12.9)a	(3.8)cde	(3.3)a	(0.50)de	(13.2)b	(0.007) bc	(1.5)bc	(1.1)a	(5.3)e	(0.17)bc
A. caeserea	137.5	18.4	736.9	7.8	502.6	0.05	48.4	33.9	96.4	2.84
A. caeserea	(11.5)a	(1.4)abc	(51.06)e	(0.73)d	(43.7)e	(0.0036)a	(4.26)e	(2.6)ef	(7.5)f	(0.15)ab
C. cibarius	106.3	12.2	673.2	4.6	588.5	0.37	35.1	31.2	49.4	3.18
C. cibarius	(8.1)a	(1.2)a	(44.7)de	(0.37)b	(65.4)f	(0.027)d	(3.7)d	(2.6)de	(3.3)bc	(0.19)c
С.	151.6	56.1	935.5	13.7	255.3	1.64	58.4	40.5	91.5	3.08
cornucopioides	(13.46)a	(4.9)f	(53.13)g	(1.22)h	(25.04)c	(0.140)f	(8.78)f	(3.2)g	(6.9)f	(0.16)bc
E hanatis	162.3	25.9	292.7	2.2	180.5	0.24	12.7	24.5	41.8	3.22
F. hepatica	(12.33)a	(1.8)de	(9.04)b	(0.15)a	(12.51)b	(0.014)c	(1.11)b	(1.2)bc	(2.4)ab	(0.17)c
Maiaantawa	176.1	20.5	578.3	6.4	367.1	0.49	20.4	45.1	60.3	2.98
M. giganteus	(14.3)a	(1.7)bcd	(35.2)c	(0.61)c	(31.3)d	(0.040)e	(2.6)c	(3.5)h	(4.8)d	(0.14)bc

Table 3. Trace element contents of mushrooms (mg/kg, dry weight)

A, B, C, D: The code names of the firms

^aMeans having the same letter(s) are not significantly different (p>0.05) by Duncan's multiple range test.

The standard deviation value is given in parentheses.

Al is reported as highly neurotoxic. Its high contents inhibit prenatal and postnatal development of the brain in humans and experimental animals [15]. In healthy humans the body burden of Al subjects has been reported to be approximately 30-50 mg/ kg body weight [16]. The highest Al content was found as 78.8 mg/kg in *A. bisporus* (C). These results may indicate that the growing medium of mushroom affects the aluminum content of mushroom highly and it can be considered that aluminum contents of the growing medium of *A. bisporus* (C) are higher than other substrates. In the literature, Al contents of some mushrooms samples ranged from 14 to 123 mg/kg [17] Al contents of mushrooms are in agreement with the literature data and the other cultivated *A. bisporus* results (14±6.8 mg/kg [17])

Ca intake is one of the most important factors that influence peak bone mass. The low bone mass is connected to the increased extent of osteoporotic fractures [18]. In most of the mushrooms that were analyzed in the study, Ca contents were generally found satisfactory. The highest Ca content was seen in *C. cornucopioides* mushroom (935.5 mg/kg).

Mn is an essential nutrient, but it can be toxic at high levels ([19]. In literature, the reported manganese values for mushrooms ranged from 2 to 100 mg/kg [20]. Also, in a wild *A. bisporus*; the Mn level was determined as 34.65 ± 2.40 mg/kg [21] This ratio was higher than that of fresh *A. bisporus* data (5.70 ± 0.41 mg/kg dw) [22]. In this study, the highest Mn was found in *C. cornucopioides* as 13.7 ± 1.22 mg/kg.

It is reported that adequate Fe is very important in order to decrease the anemia [23] and meet the iron requirement of vegetarian people. The highest Fe was found in A. caeserea (502.6 ± 43.7 mg/kg). Fe contents in some wild mushroom species have been reported in the range of 31.3-1190 mg/kg [24].

Co is a substantial trace element. However; it can be either toxic or essential for living organisms depending on its concentration level [25]. Co quantities have been noted in the ranges between 0.07 and 7.49 mg/kg, in the literature [26]. In this study, Co contents of mushrooms were found very close to each other, except F. hepatica, and in agreement with those reported in the literature.

Ni is a healthful activator for some enzyme species [25], but it can be quite toxic especially at levels exceeding the upper limit. In our study, the highest Ni was found as 58.4 ± 8.78 mg/kg in C. cornucopioides. Researchers reported that the amount of Ni in some mushroom species ranged from 8.2 to 26.7 mg/kg [27]. Our Ni content data of mushrooms, especially wild, was found higher than that in literature data.

Cu is an essential element for iron digestion and hemoglobin synthesis [28]. In an investigation, Cu contents in the mushroom species were reported between 5.11 and 92.5 mg/kg [29] In this study, Cu contents of cultivated mushroom were found between the ranges from 15.0 to 45.1 mg/kg.

Zn is one of the fundamental trace elements necessary for enzymatic functions. It plays a key role as a cofactor of more than 300 metalloenzymes [30]. It is known that mushrooms are Zn accumulators [26]. In this paper; the lowest and the highest Zn contents were 39.8 mg/kg in A. bisporus (A) and 96.0 mg/kg in A. caeserea, respectively. In the literature, Zn concentrations of some mushroom samples have been reported to be in the range of 25-122 mg/kg [31].

Se is vital for human health and necessary in the biosynthesis of important selenoenzymes [32]. The highest Se content was found in A. bisporus (A), (4.54 mg/kg). Wild and cultivated mushroom Se content values were close to each other. Data on Se content range from 0.08 to 24.7 mg/kg according to previous studies [33].

The toxic heavy metal and lead isotopes contents of cultivated mushrooms are presented in Table 4.

			r i i i i i i i i i i i i i i i i i i i		, (8,8,) .	8 9
	Pb-206	Pb-207	Pb-208	Cr	As	Cd
A 1:	0.090	0.050	0.070	0.033	0.029	0.130
A. bisporus (A)	(0.005)a	(0.003)a	(0.004)b	(0.002)bc	(0.003)a	(0.005)a
A_{1}	0.080	0.070	0.060	0.024	0.020	0.320
A. bisporus (B)	(0.006)a	(0.005)ab	(0.003)ab	(0.001)a	(0.002)a	(0.031)cd
A bigmony (C)	0.070	0.120	0.050	0.025	0.025	0.130
A. bisporus (C)	(0.003)a	(0.005)cc	(0.002)a	(0.001)a	(0.002)a	(0.005)a
A hisnomus (D)	0.130	0.060	0.120	0.030	0.022	0.110
A. bisporus (D)	(0.007)b	(0.003)ab	(0.007)d	(0.002)b	(0.003)a	(0.003)a
Destauration	0.080	0.080	0.090	0.022	0.018	0.300
P. ostreatus	(0.006)a	(0.007)b	(0.007)c	(0.002)a	(0.001)a	(0.028)c
1	0.175	0.158	0.166	0.060	0.200	0.340
A. caeserea	(0.0142)c	(0.0140)d	(0.014)e	(0.0013)e	(0.018)b	(0.029)d
C silesies	0.185	0.118	0.124	0.074	0.200	0.310
C. cibarius	(0.0164)c	(0.0113)c	(0.011)d	(0.0021)f	(0.02)b	(0.015)cd
С.	0.303	0.273	0.286	0.142	0.250	0.400
cornucopioides	(0.024)d	(0.0220)f	(0.0229)g	(0.0067)g	(0.03)c	(0.021)e
E handian	0.200	0.068	0.201	0.036	0.210	0.600
F. hepatica	(0.011)c	(0.0030)ab	(0.012)f	(0.0018)c	(0.02)b	(0.032)f
M : .	0.318	0.226	0.096	0.045	0.310	0.240
M. giganteus	(0.031)d	(0.0201)e	(0.009)c	(0.0020)d	(0.03)d	(0.021)b

Table 4. Toxic heavy metal contents and lead isotopes of mushrooms (mg/kg, dry weight)

^aMeans having the same letter(s) are not significantly different (p>0.05) by Duncan's multiple range test.

The standard deviation value is given in parentheses.

²⁰⁶Pb is the end of the decay chain of ²³⁸U, the uranium series or radium series. ²⁰⁷Pb is the final step of the Actinium series from ²³⁵U. ²⁰⁸Pb is the end of the Thorium series from ²³²Th [34]. In the literature, it is the first time that lead isotopes of wild and cultivated mushrooms were investigated and it can be said that the values of mushrooms' lead isotopes are in a safe range.

Cr is necessary for insulin activity [35] but accumulated Cr in the human body makes opposite health effects such as male reproductive system problems, respiratory, stomach, intestine and lung cancer [16] Our Cr content of mushrooms (0.024-0.142 mg/kg) was found lower than that in some literature data (1.3-24.3 mg/kg) [36].

Cd accumulates in human body and has a long biological half-life. It may cause kidney dysfunction, skeletal damage, and reproductive defects [37]. It was reported that the ranges of Cd in some mushroom species were 0.26-2.40 mg/kg [38]. Since 1999, the limits of 2.00 mg/kg and dry matter have been valid in the Czech Republic for cadmium, in wild-growing mushrooms, while 1.00 mg/kg dry matter are established for cultivated ones [39]. As can be seen in Table 4, the highest toxic element was observed in cadmium metal. However, the highest of Cd content was found as 0.60 mg/kg in *F. hepatica* and within the literature data.

Arsenic is one of the elements present in concentrations which raise chemical and toxicological questions [40]. As expected, wild mushrooms' arsenic contents were found higher than the arsenic content of cultivated mushrooms. Our arsenic values (0.018-0.31 mg/kg) of mushrooms were found lower than literature data (0.28–86 mg/kg) [41].

According to the results, the elemental composition of mushrooms was found generally significantly different (p<0.05) from each other based on Duncan's multiple range test. This situation can be attributed to many reasons. Because of the absorbing and keeping ability of some heavy metals and nutrients minerals from the medium, the accumulative capability of mushrooms can be affected by many factors such as temperature, humidity, pH, the amount of organic

material, chemical structure of the substrate, mushroom species, the development stage of mushrooms and their mycelium age. According to previous studies, elemental structure in mushroom species is generally specific [39, 42-44]. They also noted that the substances in the substrate are an important factor for the heavy metal content in mushrooms.

Correlation coefficients among the metals are presented in Table 5. The results showed that there was a strong positive correlation between Co-Mn and Cr-Ni (r = 0.843, 0.864). In addition, there was a strong negative correlation between Se-Ca and Se-Cu (r = -0.404, -0.398).

	Mg	Al	Ca	Mn	Fe	Co	Ni	Cu	Zn	Se	Pb ²⁰⁶	Pb ²⁰⁷	Pb ²⁰⁸	Cr	As	Cd
Mg	1.000															
Al	-0.163	1.000														
Ca	-0.052	0.370	1.000													
Mn	-0.178	0.270	0.347	1.000												
Fe	0.245	-0.394	ⁿ 0.132	-0.052	1.000											
Co	-0.057	0.479	0.427	0.843 ^s	-0.131	1.000										
Ni	0.031	0.175	0.465	0.607	0.546	0.599	1.000									
Cu	-0.132	0.248	0.717 ^s	0.298	0.104	0.338	0.282	1.000								
Zn	-0.148	0.154	0.312	0.567	0.224	0.369	0.766 ^s	0.229	1.000							
Se	-0.009	0.020	-0.404	ⁿ 0.158	0.168	0.122	0.000	-0.398	ⁿ -0.358 ⁱ	ⁿ 1.000						
Pb ²⁰⁶	-0.155	0.103	0.486	0.743 ^s	0.188	0.646	0.465	0.589	0.217	0.274	1.000					
Pb ²⁰⁷	-0.130	0.510	0.627	0.787 ^s	0.161	0.720 ^s	0.697	0.670	0.653	-0.099	0.700	1.000				
Pb ²⁰⁸	-0.133	0.223	0.322	0.557	-0.050	0.648	0.649	-0.046	0.424	0.138	0.346	0.294	1.000			
Cr	0.055	0.318	0.555	0.736	0.272	0.874	0.864 ^s	0.312	0.505	0.108	0.628	0.702	0.761	1.000		
As	-0.022	0.020	0.347	0.368	0.623	0.325	0.632	0.574	0.282	0.293	0.708 ^s	.0574	0.226	0.513	1.000	
Cd	-0.043	-0.108	-0.308	-0.156	-0.120	0.030	0.148	-0.264	0.149	-0.078	-0.287	-0.315	0.453	0.068	-0.029	1.00

Table 5.	Correlation	coefficients	among the metals

^s: There is strong positive correlation

ⁿ: There is strong negative correlation

4. CONCLUSION

In this study; some trace elements (Mg, Al, Ca, Mn, Fe, Co, Ni, Cu, Zn, Se), toxic heavy metal contents (Cr, As, Cd) and lead isotopes (²⁰⁶Pb, ²⁰⁷Pb and ²⁰⁸Pb) were determined in wild edible (A. caeserea, C. cibarius, C. cornucopioides, F. hepatica, M. giganteus) and cultivated mushroom (A. bisporus, P. ostreatus) species. The trace elements and toxic heavy metal contents were lower than the upper limits in each mushroom species. The elemental composition of the wild and cultivated mushrooms was statistically different (p<0.05) from each other. C. *cornucopioides* had drawn attention with the highest Ca, Mn, Ni, Cu, Zn, ²⁰⁸Pb and Cr contents. The metal accumulation in wild mushrooms was higher than cultivation mushrooms except Al and Se. The chemicals, used for hygiene in the commercial production companies during the process, did not lead to a danger for the human body. All investigated parameters revealed that there was not any health risk associated with the consumption of the analyzed wild and cultivation mushrooms.

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