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Evaluation of Macro- and Microelement Levels in Black Tea in View of Its Geographical Origin

Justyna Brzezicha-Cirocka[1] · **Małgorzata Grembecka**[1] · **Tomasz Ciesielski**[2] · **Trond Peder Flaten**[3] · **Piotr Szefer**[1]

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Abstract The aim of this study was to evaluate the elemental composition of black tea samples and their infusions in view of their geographical origin. In total, 14 elements were analyzed, 13 (Ca, K, Mg, Na, Mn, Fe, Zn, Cu, Cr, Ni, Co, Cd, and Pb) by flame atomic absorption spectrometry, and P by UV-Vis spectrometry, after mineralization of samples. It was found that K was the most abundant macroelement in the analyzed samples, whereas among microelements, the highest concentration was found for Mn. Based on the obtained data, the percentage of elements leached into the infusions as well as the daily elemental intake from tea were calculated. The daily intake from tea was compared to the recommended daily allowances (RDAs), and the highest percentages of the RDAs were found for Mn (15 %) and Co (10 %). To study the relations between elemental composition and country of origin of samples, factor analysis and cluster analysis were applied. These multivariate techniques proved to be efficient tools able to differentiate samples according to their provenance as well as plantation within the common regions.

Keywords Factor analysis · Cluster analysis · RDA · PTWI · Black tea · FAAS

Introduction

According to the Food and Agriculture Organization of the United Nations Statistics Division (FAOSTAT), total tea production was estimated at 5.3 million tons in 2011, which makes it the most commonly consumed beverage around the world. Tea production in Europe reaches 513,000 tons per year [1]. The main world tea producers are China and India (1.4 and 0.92 million tons, respectively) [2], whereas in Europe, the Russian Federation and the UK (184,000 tons and 129,000 tons, respectively) are the leading producers [1]. The countries with the highest rate of tea consumption include Paraguay and Afghanistan, 7.93 and 4.55 kg/year/person, respectively [2]. According to FAOSTAT data, Malta has the largest tea consumption per capita in Europe amounting to 2.24 kg/year [1], followed by the UK and Ireland, whereas Poland is in seventh place with 0.92 kg/year [1]. According to Polish literature, the average Polish resident consumes 2–3 cups of tea per day, and 20 % of the population consumes 4–5 cups per day [3]. Many studies have reported positive effects of tea consumption on human health, such as cancer prevention, diabetes management by reducing glucose and cholesterol levels in blood, and improved immune defense [4]. Most of the tea world consumption is black tea (80 %), whereas the remaining 20 % belong to green, oolong, red, and yellow types of this beverage. The price of tea products as well as consumer interests are usually connected with the certified geographical origin [5]. Therefore, it is important to have tools that are able to ensure good quality of products, which is usually associated with their geographical provenance. There are several studies of authentication of tea origin, using multivariate chemometric techniques, such as principal component analysis (PCA), factor analysis (FA), cluster analysis (CA), or linear discriminant analysis (LDA) [5]. These techniques have been applied to elemental and organic

✉ Piotr Szefer
pszef@gumed.edu.pl

[1] Department of Food Sciences, Medical University of Gdańsk, Al. Gen. J. Hallera 107, 80-416 Gdańsk, PL, Poland

[2] Department of Biology, Norwegian University of Science and Technology, NO-7491 Trondheim, Norway

[3] Department of Chemistry, Norwegian University of Science and Technology, NO-7491 Trondheim, Norway

composition data, obtained using various analytical techniques including HPLC [6–8], GC-MS [9–11], ^{1}HNMR [12], FT-NIRS [13], ICP-MS [5, 14–16], and FAAS [17–20]. Our previous studies have showed that FA and CA are efficient tools of green, fruit, and Pu-erh tea diversification [19, 20]. These two multivariate techniques applied to green tea data enabled differentiation of samples according to their provenance [19]. Moreover, it was also possible to diversify Pu-erh tea according to its type of confection, whereas fruit teas were differentiated according to their type [20]. Such information is of great importance to consumers, who expect that price is an equivalent of good quality products.

Therefore, in this study, we aimed to verify the geographical origin of commercialized black tea samples applying factor analysis (FA) and cluster analysis (CA) to their elemental composition. These techniques were found useful to determine the country of origin of the tea as well as its provenance within a single country. In addition, the percentage of elements leached into the tea infusions was determined, and the daily elemental intake from tea was calculated and compared to the recommended daily allowances (RDAs). For Cd and Pb, the intake from tea was compared to the provisional tolerable weekly intake (PTWI).

Materials and Methods

Samples

The analyzed tea samples (loose form and tea bags) were purchased from markets and tea shops (original tea) in Poland, and analyzed for their content of 14 elements: K, Na, Ca, Mg, Mn, Zn, Cu, Fe, P, Co, Ni, Cr, Cd, and Pb. In total, 118 types of black tea from different producers were tested, i.e., 708 analytical samples of black tea leaves and their infusions were prepared. Of the 118 tea types analyzed, 43 were purchased in tea shops (12 Chinese, 17 Indian, 10 Ceylon, and 4 Kenyan teas), and 75 in markets (loose form and tea bags) (Table 1).

Preparation of Samples and Elemental Analysis

The bulk teas were homogenized and representative samples were mineralized in an electric furnace and then analyzed by flame atomic absorption spectrometry (FAAS) according to the previously published procedure by Brzezicha-Cirocka et al. [19, 20].

Method Validation

The limit of detection (LOD) and limit of quantification (LOQ) for all the analyzed elements were calculated based on the independently prepared blank samples measurements. According to the method described by Konieczka and Namieśnik [21], LODs were set to blank means +3SD, where blank mean is a result of all blank samples measurements and SD is their standard deviation; whereas LOQ was calculated by multiplying LOD by a factor of three. Data for the validation procedure are given in Table 2.

The reliability of the method was determined using certified reference materials, i.e., Oriental Basma Tobacco Leaves (INCT-OBTL-5) and Polish Virginia Tobacco Leaves (INCT-PVTL-6). They were prepared according to the same procedure as the analytical samples. Recoveries of the studied elements ranged between 87 and 113 % (RSDs between 0.02–10.3 %) of the certified values for all the elements (Table 3).

Statistical Analysis

The Shapiro-Wilk test showed that the data were not normally distributed; therefore, nonparametric tests were applied [22]. Moreover, data standardization was adopted, and the correlation analysis was performed using Spearman rank analysis. Kruskal-Wallis test, factor analysis, and cluster analysis were conducted in order to obtain statistically significant information about the quality and origin of samples.

Results and Discussion

Macroelements

The highest contents of Ca and Na were found in marketed teas (264 and 89.3 mg/100 g, respectively), whereas Indian and Kenyan teas were characterized by the lowest Na level (30.7 and 21.8 mg/100 g, respectively). Products from China, India, Ceylon, and Kenya had similar amounts of Ca (153–168 mg/100 g). Comparable Ca results (215 mg/100 g) were obtained by Dambiec et al. [23], whereas similar Na levels (88 mg/100 g) were reported by Soomro et al. [24] and Yemane et al. [25]. Among all macroelements, the highest levels were found for K (2349–2981 mg/100 g). Considerable variation was found in the content of Mg, as the highest levels amounted to 822 mg/100 g in Indian teas, and the lowest to 518 mg/100 g in marketed teas (Table 4). The latter value is higher than the one obtained by Gerbresadik and Chandravanshi [26] (354 mg/100 g). Indian, Ceylon, and Kenyan products contained similar amounts of P (305–359 mg/100 g), and Chinese tea had the highest P concentration (408 mg/100 g). Malik et al. [27] published comparable results for P (366 mg/100 g). Dambiec et al. [23] estimated a percentage of Na leaching (45.3 %) similar to ours (40.0 %), while Szymczycha-Madeja et al. [17] classified this macroelement as highly extractable (>55 %), which was confirmed in the case of teas from Kenya (62.0 %). The percentages of Ca leaching were the highest in Chinese teas (17.2 %) and the

Table 1 Characteristics of the analyzed products

No.	Name of tea	Producer	Country/producer declaration	Confection
Original tea				
1.	Ceylon FBOPF “Malatiyana”	Maraska	Ceylon	Loose
2.	Ceylon OP “Lumbini”	Maraska	Ceylon	Loose
3.	Ceylon “Kendy”	Maraska	Ceylon	Loose
4.	Ceylon UVA OPI “Ivy Hills”	Maraska	Ceylon	Loose
5.	Ceylon Dimbula	Five o’clock	Ceylon	Loose
6.	Ceylon Raigama Korales	Five o’clock	Ceylon	Loose
7.	Ceylon Pothotuwa	Five o’clock	Ceylon	Loose
8.	Ceylon Sithaka FBOPFEXS W	Five o’clock	Ceylon	Loose
9.	Ceylon Earl Grey	Five o’clock	Ceylon	Loose
10.	Ceylon High Grown	Time to tea	Ceylon	Loose
11.	Yunnan Golden OP	Maraska	China	Loose
12.	Golden Monkey	Maraska	China	Loose
13.	China OP Keemun	Maraska	China	Loose
14.	Yunnan Special Black	Five o’clock	China	Loose
15.	China Keemun Mao Feng	Five o’clock	China	Loose
16.	Golden Yunnan[a]	Five o’clock	China	Loose
17.	Lapsang Souchong[a]	Five o’clock	China	Loose
18.	China Black Golden Monkey	Five o’clock	China	Loose
19.	Lapsang Souchong[a]	Time to tea	China	Loose
20.	China Keemun Congu	Time to tea	China	Loose
21.	Yunnan Black Premium	Time to tea	China	Loose
22.	Golden Yunnan[a]	Time to tea	China	Loose
23.	Darjeeling FTGFOPI “Himalaya”	Maraska	India	Loose
24.	Assam TGFOPI “Dikom”	Maraska	India	Loose
25.	Assam TGFOP “Ambaguri”	Maraska	India	Loose
26.	Assam Jamguri FTGFOP1	Five o’clock	India	Loose
27.	Assam Dagapur	Five o’clock	India	Loose
28.	Darjeeling Castleton 2011 WP	Five o’clock	India	Loose
29.	Assam Satishpur TGFOP W	Five o’clock	India	Loose
30.	Assam Marangi FTGFOP1	Five o’clock	India	Loose
31.	Darjeeling Thurbo FTGFOP	Five o’clock	India	Loose
32.	Assam Halmari GTGFOP CL W	Five o ‘clock	India	Loose
33.	Darjeeling Margarets Hope	Five o’clock	India	Loose
34.	Assam Dikom	Time to tea	India	Loose
35.	Assam Sec. Flush	Time to tea	India	Loose
36.	Assam Dekorai	Time to tea	India	Loose
37.	Darjeeling Gielle	Time to tea	India	Loose
38.	Darjeeling First Flush	Time to tea	India	Loose
39.	Darjeeling Sec. Flush	Time to tea	India	Loose
40.	Kenia GFOP “Milima	Maraska	Kenia	Loose
41.	Kenia TGFOP Golden Tipped	Maraska	Kenia	Loose
42.	Kenia Marinyn	Five o’clock	Kenia	Loose
43.	Ruanda Rukeri	Five o’clock	Kenia	Loose
Marketed tea				
1.	English Breakfast[a]	Ahmad Tea	Ceylon	Loose
2.	Ceylon Tea	Ahmad Tea	Ceylon	Loose
3.	Earl Grey Tea[a]	Ahmad Tea	Ceylon	Loose
4.	Earl Grey[a]	Ahmad Tea	Ceylon	Bags

Table 1 (continued)

No.	Name of tea	Producer	Country/producer declaration	Confection
5.	English Breakfast[a]	Ahmad Tea	Ceylon	Bags
6.	English No.1	Ahmad Tea	Ceylon	Bags
7.	Assam[a]	Ahmad Tea	Ceylon	Bags
8.	Ceylon	Ahmad Tea	Ceylon	Bags
9.	English V.I.P Tea	Brizton	Ceylon	Bags
10.	English Royal Tea[a]	Chelton Tea Collection	Ceylon	Loose
11.	Scottish Breakfast	Chelton Tea Collection	Ceylon	Loose
12.	English Royal Tea[a]	Chelton Tea Collection	Ceylon	Bags
13.	Ceylon Supreme Tea	Dilmah	Ceylon	Loose
14.	Earl Grey Tea[a]	Dilmah	Ceylon	Loose
15.	Meda Watte	Dilmah	Ceylon	Loose
16.	Ran Watte	Dilmah	Ceylon	Loose
17.	Uda Watte	Dilmah	Ceylon	Loose
18.	English Breakfast Tea	Dilmah	Ceylon	Loose
19.	English Afternoon Tea	Dilmah	Ceylon	Bags
20.	Premium Tea	Dilmah	Ceylon	Bags
21.	Ceylon Gold	Dilmah	Ceylon	Bags
22.	Perfect Ceylon Tea	Dilmah	Ceylon	Bags
23.	Elegant Earl Grey	Dilmah	Ceylon	Bags
24.	Ceylon OP	Drury	Ceylon	Loose
25.	Royal Ceylan	Lipton	Ceylon	Loose
26.	Yellow Label Tea[a]	Lipton	Ceylon	Bags
27.	Gold Tea Black	Lipton	Ceylon	Bags
28.	Mild Ceylon	Lipton	Ceylon	Bags
29.	Earl Grey[a]	Sir Roger	Ceylon	Bags
30.	Ceylon Gold	Sir William’s	Ceylon	Bags
31.	Black Tea Ceylon	Sir William’s	Ceylon	Bags
32.	Super Pekoe	Tarlton	Ceylon	Loose
33.	Ceylon Orange Pekoe Tea	Twinings	Ceylon	Bags
34.	Yunnan	Loyd Tea	China	Loose
35.	Prince of Wales	Twinings	China	Bags
36.	Black Tea[a]	Yunnan	China	Loose
37.	Darjeeling[a]	Ahmad Tea	India	Loose
38.	Darjeeling[a]	Ahmad Tea	India	Loose
39.	Assam[a]	Ahmad Tea	India	Loose
40.	Darjeeling[a]	Darvilles of Windsor	India	Loose
41.	Royalty Assam	Darvilles of Windsor	India	Loose
42.	Maharajah Reseve Assam	Dilmah	India	Bag
43.	Rich Assam	Lipton	India	Bags
44.	Darjeeling[a]	Premier’s Tea Limited	India	Loose
45.	Earl Grey[a]	Premier’s Tea Limited	India	Loose
46.	Darjeeling SFTGFOP1	Rich Mont	India	Loose
47.	Earl Grey[a]	Sir William’s	India	Bags
48.	Intensive Tea	Tetley	India	Bags
49.	Darjeeling Tea[a]	Twinings	India	Loose
50.	Darjeeling Tea[a]	Twinings	India	Bags
51.	English Tea No.1	Ahmad Tea	–	Loose
52.	English Breakfast[a]	Time to tea	–	Loose
53.	Earl Grey[a]	Time to tea	–	Loose

Table 1 (continued)

No.	Name of tea	Producer	Country/producer declaration	Confection
54.	English Breakfast[a]	Darvilles of Windsor	–	Loose
55.	Earl Grey[a]	Darvilles of Windsor	–	Loose
56.	Earl Grey[a]	Dilmah	–	Bags
57.	Daily Superior	Irving	–	Loose
58.	Daily Classic	Irving	–	Bags
59.	Earl Grey[a]	Irving	–	Bags
60.	Russian Earl Grey	Lipton	–	Loose
61.	Yellow Label Tea[a]	Lipton	–	Loose
62.	Earl Grey Classic	Lipton	–	Bags
63.	Taste of London	Lipton	–	Bags
64.	Earl Grey[a]	Loyd Tea	–	Loose
65.	Fairtrade Luxury Gold Tea	Marks & Spancer	–	Bags
66.	Earl Grey[a]	Maraska	–	Loose
67.	Black Tea[a]	Minutka	–	Bags
68.	Black Tea[a]	Saga	–	Loose
69.	Black Tea[a]	Saga	–	Bags
70.	Earl Grey[a]	Saga	–	Bags
71.	English Breakfast[a]	Twinings	–	Loose
72.	Earl Grey[a]	Twinings	–	Loose
73.	Prince of Wales Tea	Twinings	–	Loose
74.	Earl Grey[a]	Twinings	–	Bags
75.	Simply Tea	Twinings	–	Bags

[a] Various producer or confections of tea the same name

lowest in Kenyan teas (5.89 %). The lowest percentage of Mg leaching was found in Chinese teas (29.6 %), which is comparable to findings reported by Dambiec et al. [23]. Magnesium and P leaching percentages ranged between 29.6–

Table 2 Results of the validation procedure of the analytical methodology

Element	Linearity			LOD (mg/100 g)	LOQ (mg/100 g)
	Calibration curve range (μg/mL)	Calibration curve	R^2		
Ca	2.00–15.0	$y = 0.05801\times + 0.0089$	0.999	0.020	0.060
K	0.50–1.50	$y = 0.00048\times + 0.0149$	0.997	0.040	0.120
Mg	0.10–0.90	$y = 0.00107\times + 0.0220$	0.998	0.020	0.060
Na	0.50–1.20	$y = 0.00071\times + 0.0192$	0.996	0.020	0.060
P	0.10–1.20	$y = 0.00444\times + 0.0117$	0.999	0.030	0.090
Mn	0.15–5.00	$y = 0.00015\times + 0.0058$	0.999	0.020	0.060
Fe	1.00–10.0	$y = 0.00006\times + 0.0082$	0.996	0.010	0.030
Zn	0.20–1.50	$y = 0.00034\times + 0.0079$	0.998	0.020	0.060
Cu	0.50–4.00	$y = 0.00013\times + 0.0022$	0.999	0.009	0.027
Co	1.00–5.00	$y = 0.00008\times + 0.0052$	0.999	0.003	0.009
Cd	0.20–2.00	$y = 0.00035\times + 0.0040$	0.999	0.003	0.009
Cr	0.20–2.00	$y = 0.00005\times + 0.0007$	0.999	0.001	0.003
Ni	0.50–2.00	$y = 0.00008\times + 0.0007$	0.999	0.002	0.006
Pb	0.20–2.00	$y = 0.00004\times + 0.0004$	0.999	0.004	0.012

Table 3 Element concentrations and RSD with recovery data for the certified reference materials Oriental Basma Tobacco Leaves (INCT-OBTL-5) and Polish Virginia Tobacco Leaves (INCT-PVTL-6)

Element	Certified values (mg/100 g)	Determined values (mg/100 g)	RSD (%)	Recovery (%)
Ca[a]	3859 ± 142	3566 ± 149	4.17	92
Ca[b]	2297 ± 78	2513 ± 13.4	0.50	109
Co[a]	0.10 ± 0.007	0.09 ± 0.005	5.65	90
Cu[a]	1.01 ± 0.04	1.00 ± 0.01	1.42	99
Cu[b]	0.51 ± 0.02	0.54 ± 0.01	2.00	106
Cd[a]	0.26 ± 0.01	0.28 ± 0.01	3.48	105
Cd[b]	0.22 ± 0.01	0.21 ± 0.001	0.40	95
Cr[a]	0.63[c]	0.56 ± 0.0001	0.02	89
Cr[b]	0.09[c]	0.09 ± 0.001	1.30	98
Mg[a]	853 ± 34	845 ± 4.96	0.59	99
Mg[b]	241 ± 9	247 ± 6.01	2.40	102
Mn[a]	18.0 ± 0.6	20.2 ± 0.34	1.67	112
Mn[b]	13.6 ± 0.5	15.3 ± 0.18	1.20	113
Zn[a]	5.24 ± 0.18	5.59 ± 0.24	4.31	107
Zn[b]	4.36 ± 0.1	4.64 ± 0.01	0.30	106
K[a]	2271 ± 76	2449 ± 50.3	2.06	108
K[b]	2640 ± 90	2692 ± 31.6	1.20	102
Na[b]	6.24[c]	5.50 ± 0.03	0.50	88
Pb[a]	0.20 ± 0.03	0.19 ± 0.01	5.60	95
Pb[b]	0.10 ± 0.01	0.09 ± 0.01	10.3	89
P[a]	170 ± 12	170 ± 0.41	0.24	100
P[b]	242 ± 5	239 ± 0.96	0.40	99
Ni[a]	0.85 ± 0.05	0.80 ± 0.078	9.80	94
Ni[b]	0.15 ± 0.01	0.13 ± 0.001	0.6	87
Fe[a]	149[c]	160 ± 1.01	0.63	107
Fe[b]	25.8[c]	28 ± 0.30	1.1	109

[a] Oriental Basma Tobacco Leaves INCT-OBTL-5

[b] Polish Virginia Tobacco Leaves INCT-PVTL-6

[c] Information value

39.7 and 26.8–38.2 %, respectively. The average percentage of Mg extraction to infusions (35.3 %) obtained by Dambiec et al. [23] and Gallaher et al. [28] is comparable to our results.

Microelements

Manganese was the microelement found in the highest concentrations in our study, and the highest levels were determined in Kenyan and marketed teas (53.2 and 56.3 mg/100 g, respectively). Shaltout and Abd-Elkader [29] reported a slightly higher average Mn content (61.8 mg/100 g). Chinese samples had the highest Fe and Zn content (0.90 and 4.25 mg/100 g, respectively). According to Mupenzi et al. [30], low Fe concentration can be induced by high Mn levels. What is more, high Fe levels can cause Mn deficiency in tea plants. The determined amounts of Zn are comparable to those obtained by Al-Oud [31] for Chinese and Indian teas (2.67–5.39 mg/100 g). Copper levels in samples from Ceylon and Kenya (1.83 and 1.85 mg/100 g) were comparable and slightly lower than in Chinese, Indian, and marketed teas (2.25, 2.39, and 2.28 mg/100 g, respectively). Similar Cu results were reported by McKenzie et al. [32] (1.70 mg/100 g). Cobalt levels, which amounted to 0.02 mg/100 g, were comparable to those reported by Shaltout and Abd-Elkader [29]. Chinese and Indian samples had similar amounts of Cr (0.08 and 0.07 mg/100 g) and Ni (0.50 and 0.58 mg/100 g). Similar results were obtained by Shaltout and Abd-Elkader [29] for Ni (0.61 mg/100 g) and by Barone et al. [4] for Cr (0.04 mg/100 g). Barone et al. [4] reported in their study that Chinese tea samples generally had higher levels of elements than those originating from India.

The lowest percentage of leaching of Mn (25.1 %) was found in marketed teas and the highest (44.8 %) in Indian teas. For Fe and Zn, percentages of leaching showed similar levels in all the analyzed tea samples (29.1–38.7 and 33.9–45.1 %, respectively). Among all the analyzed elements, Ni showed the highest percentage of leaching, which ranged from 56.2 % (Chinese tea) to 75.8 % (Kenyan tea).

Table 4 Concentration of bioelements and toxic metals in dry tea samples in milligrams/100 g ($\bar{x}$ ± SD range) and percentage of leaching

Elements	China	India	Ceylon	Kenya	Marketed
n	12 × 3	17 × 3	10 × 3	4 × 3	75 × 3
Ca	161 ± 101	153 ± 35	168 ± 51	155 ± 33	264 ± 52
	(22–449)	(105–220)	(87–250)	(105–193)	(133–420)
	17.2 ± 12.0 %	11.1 ± 8.22 %	9.54 ± 6.37 %	5.89 ± 1.60 %	11.1 ± 7.06 %
K	2666 ± 223	2803 ± 161	2981 ± 153	2738 ± 119	2349 ± 271
	(2322–2992)	(2474–3084)	(2656–3207)	(2536–2838)	(1845–3057)
	28.3 ± 8.03 %	33.6 ± 4.65 %	30.6 ± 6.25 %	28.4 ± 4.27 %	23.1 ± 4.08 %
Mg	764 ± 77	822 ± 103	769 ± 41.7	774 ± 63.1	518 ± 253
	(669–936)	(601–1052)	(704–829)	(699–873)	(164–901)
	29.6 ± 8.26 %	36.5 ± 3.07 %	35.6 ± 3.59 %	39.7 ± 4.37 %	35.1 ± 11.8 %
Na	67.3 ± 61.4	30.7 ± 11.5	55.5 ± 31.0	21.8 ± 6.06	89.3 ± 126
	(24.2–267)	(17.5–60.3)	(23.9–122)	(13.4–30.5)	(10.4–728)
	29.7 ± 11.4 %	47.8 ± 18.2 %	32.6 ± 12.2 %	62.0 ± 12.7 %	27.7 ± 20.7 %
Mn	30.9 ± 11.8	30.4 ± 10.5	27.7 ± 10.3	53.2 ± 12.6	56.3 ± 28.4
	(11.7–59.1)	(12.8–49.0)	(16.4–50.0)	(33.2–68.2)	(16.1–143)
	41.4 ± 14.4 %	44.8 ± 11.3 %	39.1 ± 8.53 %	44.3 ± 9.51 %	25.1 ± 7.00 %
P	408 ± 58.0	359 ± 39.9	305 ± 52.8	344 ± 25.3	296 ± 66
	(317–482)	(309–460)	(236–434)	(310–380)	(198–490)
	34.1 ± 9.50 %	34.7 ± 8.66 %	26.8 ± 8.53 %	31.6 ± 5.35 %	38.2 ± 7.66 %
Fe	0.90 ± 0.26	0.57 ± 0.29	0.43 ± 0.09	0.53 ± 0.01	0.73 ± 0.37
	(0.37–1.35)	(0.30–1.22)	(0.30–0.63)	(0.51–0.54)	(0.23–2.50)
	38.7 ± 17.9 %	38.0 ± 26.1 %	36.7 ± 22.0 %	29.1 ± 9.62 %	37.5 ± 23.8 %
Zn	4.25 ± 0.63	3.70 ± 0.60	2.70 ± 0.52	2.74 ± 0.22	3.29 ± 0.67
	(3.26–5.72)	(2.92–5.05)	(2.11–3.91)	(2.50–2.99)	(2.04–5.22)
	34.6 ± 11.2 %	36.2 ± 10.6 %	41.3 ± 10.6 %	45.1 ± 4.11 %	33.9 ± 11.3 %
Cu	2.25 ± 0.42	2.39 ± 0.50	1.83 ± 0.14	1.85 ± 0.17	2.28 ± 0.57
	(1.55–2.91)	(1.67–3.34)	(1.62–2.07)	(1.67–2.07)	(1.26–3.98)
	15.4 ± 3.42 %	16.9 ± 2.57 %	15.0 ± 1.75 %	14.1 ± 1.91 %	18.9 ± 5.34 %
Co	0.03 ± 0.01	0.02 ± 0.01	0.02 ± 0.01	0.02 ± 0.01	0.02 ± 0.01
	(0.01–0.04)	(0.01–0.04)	(0.01–0.05)	(0.01–0.04)	(0.01–0.05)
	39.4 ± 21.3 %	40.0 ± 25.1 %	43.6 ± 20.0 %	33.9 ± 4.41 %	33.4 ± 18.7 %
Cd	0.005 ± 0.001				
	(<LOD-0.007)	<LOD	<LOD	<LOD	<LOD
	<LOD				
Cr	0.08 ± 0.02	0.07 ± 0.03	0.05 ± 0.01	0.11 ± 0.06	0.17 ± 0.16
	(0.04–0.11)	(0.04–0.12)	(0.04–0.06)	(0.04–0.18)	(0.04–1.28)
	24.1 ± 8.48 %	34.1 ± 13.4 %	28.2 ± 9.34 %	31.7 ± 6.95 %	28.1 ± 13.3 %
Ni	0.50 ± 0.21	0.58 ± 0.13	0.35 ± 0.10	0.36 ± 0.05	0.42 ± 0.15
	(0.22–1.01)	(0.28–0.85)	(0.22–0.63)	(0.29–0.44)	(0.10–0.93)
	56.2 ± 18.6 %	67.7 ± 11.3 %	68.8 ± 17.2 %	75.8 ± 17.0 %	64.0 ± 24.4 %
Pb	0.05 ± 0.04	0.02 ± 0.01	0.02 ± 0.01	0.013 ± 0.003	0.03 ± 0.04
	(0.01–0.15)	(<LOD-0.06)	(0.01–0.04)	(0.01–0.02)	(<LOD-0.32)
	33.9 ± 21.3 %	18.9 ± 22.2 %	28.7 ± 28.6 %	45.4 ± 22.4 %	27.4 ± 26.4 %

LOD for Cd = 0.003 mg/100 g; LOD for Pb = 0.004 mg/100 g

n number of samples multiplied by number of analytical subsample

Many factors influence the contents of trace metals in tea leaves. According to Milani et al. [33], variations in mineral composition of tea leaves can be explained by the age of leaves used in the production (old or young), soil composition, rainfall amount, and growing conditions in general. Unfortunately, commercial teas often have unknown geographical origin, as they are a mixture of leaves from different locations [33]. Black tea is produced through leaf fermentation in contrast to green tea, which results in higher levels of certain trace elements [34]. Regulations have been established for many vegetable products in many countries, but the European Union has no specified regulations about the acceptable metal content in tea.

Toxic Metals

In general, the analyzed samples contained more Pb than Cd. The levels of Cd varied little, i.e., 0.003–0.005 mg/100 g. Similar results for Cd were obtained by Milani et al. [33] (0.001–0.002 mg/100 g) and significantly lower ones by Barone et al. [4] (0.0004 mg Cd/100 g). Chinese teas had the highest Pb content (0.05 mg/100 g), which is comparable with the results reported by Barone et al. [4] (0.05 mg Pb/100 g). The percentage of leaching of Pb to infusions varied considerably, as the highest was found for Kenyan teas (45.4 %) and the lowest for Indian ones (18.9 %).

The contents of heavy metals in tea leaves may be a result of contamination that can be caused by many factors such as use of various manufacturing and agronomic processes and of fertilizers [29, 35, 36]. Among the main sources of Pb in the environment are leaded fuel, waste incineration, and industry [37]. Lead pollution is correlated with urbanization and population density. Moreover, higher levels of this heavy metal in tea samples can be attributed to contamination during the process of tea production and its packing [37]. Cadmium found in tea leaves might be a result of phosphate and zinc fertilizers usage [38]. It is estimated that Cd from phosphate fertilizers constitutes >50 % of the total input to agricultural land not heavily polluted or heavily industrialized [39, 40]. Sarma et al. [41] reported that heavy metal contaminations of tea leaves might be explained by the position of the tea cultivation area. In that study, an Assam plantation was situated in the vicinity of the oldest crude oil exploration station, and accidental spillage during drilling and transportation could be the main sources of tea field contamination. Moreover, it has been established that heavy metals enter into plant bodies in acidic soil, in which tea usually grows [42].

Correlation

Nonparametric Spearman's rank test was performed at three levels of significance ($p < 0.05$, $p < 0.01$, $p < 0.001$), and both positive and negative correlations were found between the analyzed elements. The most significant positive correlations ($p < 0.001$) were found for P-Zn, Mn-Co, and Mn-Cr for all the countries. There were also recorded important interelement correlations among black tea samples from Indian and Ceylon plantations. Strong interdependences ($p < 0.01$) were also found between Ca-Mn, P-Zn, Mn-Co, Zn-Ni, and Cu-Co.

Kruskal-Wallis Test

Through the Kruskal-Wallis test, it was possible to determine statistically significant differences in the analyzed database. Relationships were found between the geographical provenance of tea and concentrations of elements including the following: Na ($H = 18.596$; $p = 0.001$), P ($H = 14.533$; $p = 0.001$), Mn ($H = 9388$; $p = 0.025$), Fe ($H = 12.002$; $p = 0.007$), Zn ($H = 26.166$; $p = 0.000$), Cu ($H = 12.861$; $p = 0.005$), and Ni ($H = 11.929$; $p = 0.008$). Dunn's test was also performed, confirming the outcome of the Kruskal-Wallis test. The results of Dunn's test are shown in Table 5. Kruskal-Wallis test and Dunn's test (Table 6) were also performed for tea samples from Asia, i.e., India (plantation Assam and Darjeeling) and the island of Ceylon. Relationships were found between provenance of tea and concentrations of several elements: Na ($H = 10.739$; $p = 0.005$), P ($H = 10.992$; $p = 0.004$), Mn ($H = 7.218$; $p = 0.027$), Fe ($H = 11.833$; $p = 0.003$), Zn ($H = 14.679$; $p = 0.001$), Cu ($H = 13.443$; $p = 0.001$), Cr ($H = 7.382$; $p = 0.025$) and Ni ($H = 12.232$; $p = 0.002$).

Factor Analysis

The results of the factor analysis (FA), conducted on raw data sets of black tea obtained from tea shops, are shown in Fig. 1a and b. Factor analysis performed with all the analyzed metals did not give clear output, and thus, we decided to narrow the data set to the elements Na, Mn, Ni, Cu, Fe, P, and Zn. Factor analysis was applied to data of elements which were found significant in the Kruskal-Wallis test. The final choice of descriptors was done by a series of factor analyses, which were performed in order to verify the clarity of the outcome. As a result, two factors were obtained, i.e., F1 (35.7 % of the total variance) and F2 (23.5 % of the total variance). Both factors cumulatively explain 59.2 % of the total variance, whereas the eigenvalues for F1 and F2 are 2.14 and 1.41, respectively. As can be seen in Fig. 1a, samples from all the analyzed regions can be distinguished, i.e., Ceylon, China, India, and Kenya. The scatter plot of loadings was drawn for F1–F2 in

Table 5 Results of the post hoc Dunn's test conducted for the analyzed data matrix for tea samples from Ceylon, China, India, and Kenya. There are only given elements for which $p < 0.05$

	Ceylon	China	India	Kenya
Ceylon	–	P, Zn, Fe	Cu, Na, Ni, Zn	Na
China	P, Zn, Fe	–	Na, Fe	Na, Zn
India	Cu, Na, Ni, Zn	Na, Fe	–	
Kenya	Na	Na, Zn		–

Table 6 Results of the post hoc Dunn's test conducted for the analyzed data matrix for tea samples from India and Ceylon. There are only given elements for which $p < 0.05$

	Ceylon	Assam	Darjeeling
Ceylon	–	Na, Cu, Cr, Ni	P, Fe, Zn, Ni
Assam	Na, Cu, Cr, Ni	–	Mn, Fe
Darjeeling	P, Fe, Zn, Ni	Mn, Fe	–

order to identify elements responsible for the grouping of objects (Fig. 1b). Higher values of F1 correspond to Indian and Chinese samples, which were described by Ni, Cu, Fe, P, and Zn. Phosphorus, Fe, and Zn were responsible for differentiation of Chinese tea, whereas the highest amounts of Ni and Cu were noted in Indian samples. Such high levels of these metals in samples of these two origins could possibly be explained by product contamination during its manufacture or area pollution. Simultaneously, high amounts of P in Chinese samples may be related to the increasing use of fertilizers, which was confirmed by research conducted by Mupenzi et al. [30].

Lower values of F1 characterize Kenyan and Ceylon tea samples which are described by Mn and Na (Fig. 1a, b). Kenyan tea was differentiated by Mn as significant differences were found in Mn concentrations between these two groups of tea. Sodium was found to be a descriptor of Ceylon teas, which may be associated with geographical location of the plantation, as Ceylon is an island surrounded by the waters of the Indian Ocean. Higher values of F2 described Ceylon, Chinese, and partly Indian tea samples, which can be identified by Na, Fe, P, and Zn. Lower values of F2 were associated with Indian and Kenyan samples, which were differentiated by Mn, Ni, and Cu (Fig. 1a, b).

Factor analysis was also performed for Indian plantations, i.e., Assam and Darjeeling, and Ceylon samples in order to differentiate samples within plantations (Fig. 2a, b). It was

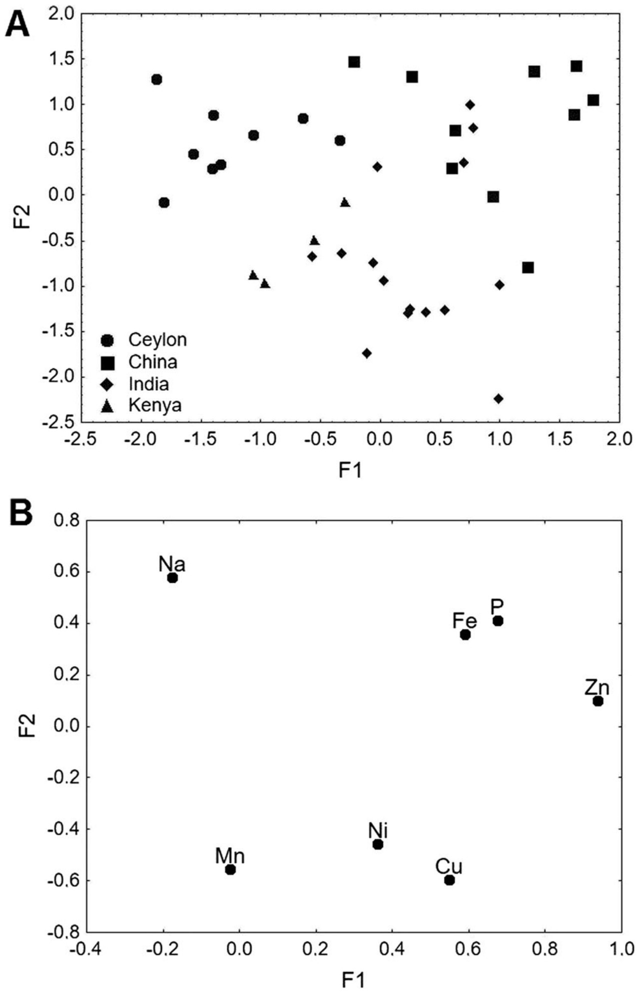

Fig. 1 **a** Scatter plot of objects samples of two factors of the all tea samples from Ceylon, China, India, and Kenya. **b** Scatter plot of loading for elements in all the analyzed tea samples from Ceylon, China, India, and Kenya

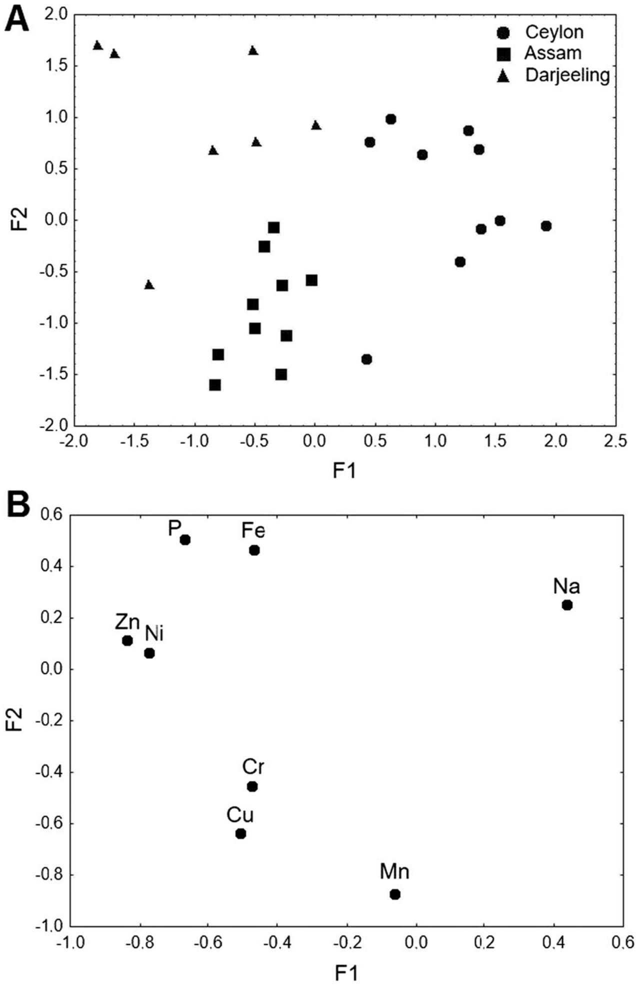

Fig. 2 **a** Scatter plot of objects samples of two factors of the all tea samples from India plantations and Ceylon. **b** Scatter plot of loadings for elements in all the analyzed tea samples from India plantations and Ceylon

found that 57.2 % of the total variance is explained by F1 (33.1 %) and F2 (24.1 %). The eigenvalues were 2.64 for F1 and 1.93 for F2. The factor analysis clearly differentiated between tea samples originating from different plantations. Ceylon samples were described by higher F1 values and Na, which might be associated with its significant content in soils of this country (Fig. 2a, b). Lower values of F1 corresponded to Assam and Darjeeling samples, which were described by P, Fe, Zn, Ni, Cr, Cu, and Mn. Tea samples from Darjeeling were differentiated by P, Fe, and Zn, whereas Assam tea samples were significantly correlated with Cr, Cu, and Mn. Factor 2 differentiated samples from India (Assam and Darjeeling) as its higher values corresponded to Darjeeling plantations and lower values to Assam ones. Although Ni is characterized with positive F2 loading, there were no significant variations in its content in Assam and Darjeeling samples.

Cluster Analysis

The cluster analysis (CA) was based on Ward's method with the usage of the Euclidean distance. Application of CA made it possible to differentiate samples according to their origin, i.e., Ceylon and Indian plantations (Fig. 3). Assam plantations were discriminated by Cr, Cu, and Mn, and Darjeeling ones by Fe, Zn, and P. Ceylon tea samples corresponded to Na, which was already confirmed to be a discriminative element for this region. However, only few samples were assigned to the improper cluster, which might be due to the similarity between samples, especially in view of their contamination with heavy metals. What is more interesting, it was noted that Darjeeling samples are more similar to Ceylon samples than to the other Indian plantation (Assam) (Fig. 3).

Thus, it can be concluded that CA similarly to FA is able to distinguish tea samples in view of their geographical origin, which might be helpful when authenticity assessment and fraud detection is necessary.

Recommended Dietary Intake

The daily intake of bioelements from tea was evaluated in view of the latest available Polish [43] and American recommended dietary intakes (RDA) [44]. Consumption of one cup of black tea (200 mL) results in intakes of Ca, K, Na, Mg, and P in the range of 0.02–1.30 % of their respective RDAs. Thus, black tea is not a rich source of these macroelements, despite the fact that their concentrations in tea leaves are the highest among all the analyzed elements (Table 7). Realization of the recommended daily intakes for microelements such as Fe, Zn, Cu, and Ni is less than 1.0 %. For Cr, intake of 200 mL of tea infusion supplies 2.8 and 4.0 % of the RDA for men and women, respectively. Black tea is a significant source of Mn and Co, one cup daily provides 15 % of the RDA for Mn and 10 % for Co. However, Mn bioavailability amounts up to 40 % [45]. Therefore, only 6 % of Mn present in one cup of black tea will be absorbed. Moreover, according to the World Health Organization [46] there is no quantitative information available to indicate toxic levels of manganese in the human diet. High Co content in tea leaves may possibly be explained by the use of Co-containing fertilizers. There is evidence that Co in higher plants promotes the formation of chlorophyll and plant growth [47]. According to Kabata-Pendias and Szteke [47], approximately 50 % of Co will be absorbed in the gastrointestinal tract. Furthermore, Co absorption can be increased among individuals who are Fe deficient.

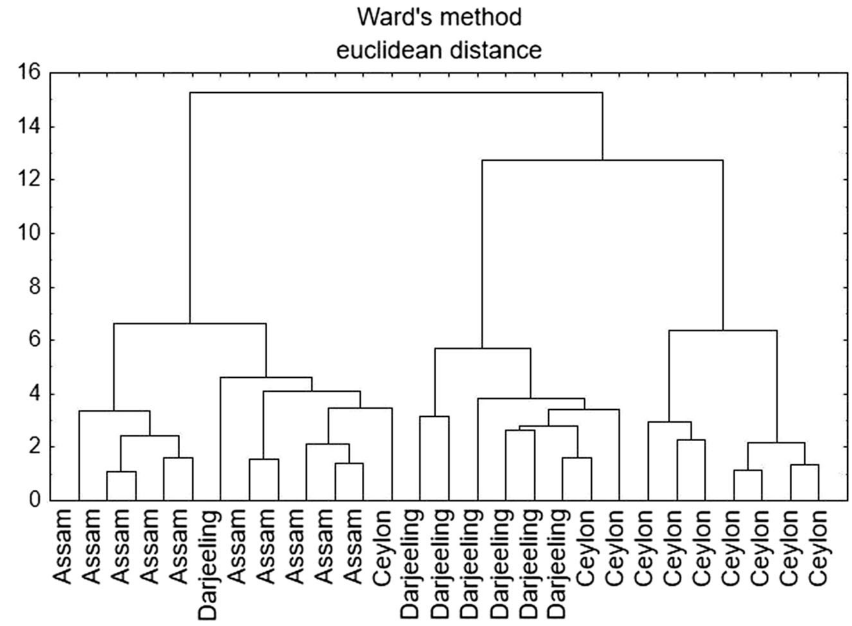

Fig. 3 Hierarchical dendrogram for tea samples from India and Ceylon plantations

Table 7 Comparison of Recommended Dietary Allowance (for a person weighing 70 kg through consumption of one cup (200 mL) of black tea beverage) to the daily intake from tea with consideration of PTWI for Pb

Element	Recommended daily allowance (RDA) (mg/day/person)		Average content in 200 mL of infusion (mg/200 mL)	Percentage of RDA	
	Male (31–50 years)	Female (31–50 years)		Male (31–50 years)	Female (31–50 years)
Ca	1000	1000	0.49 ± 0.38 0.03–1.83	0.05	0.05
K	4700	4700	13.2 ± 4.27 4.19–24.5	0.3	0.3
Mg	420	320	4.21 ± 1.99 0.46–8.34	1.0	1.3
Na	1500	1500	0.27 ± 0.12 0.07–0.67	0.02	0.02
Mn[a]	2.3	1.8	0.27 ± 0.13 0.07–0.84	11.7	15
Fe[b]	10	18	0.005 ± 0.003 0.0002–0.01	0.05	0.03
P	700	700	2.29 ± 0.69 0.98–4.54	0.3	0.3
Zn	11	8	0.02 ± 0.009 0.001–0.056	0.2	0.2
Cu	0.9	0.9	0.01 ± 0.003 0.003–0.017	1.1	1.1
Co[c]	0.002	0.002	0.0002 ± 0.0001 <LOD-0.0005	10	10
Cr[a]	0.035	0.025	0.001 ± 0.0004 0.00005–0.002	2.8	4.0
Ni[a]	1	1	0.006 ± 0.003 <LOD-0.01	0.6	0.6
Element	PTWI	PTWI for a person weighing 70 kg	The average content in 200 mL beverage (mg/200 mL)	Realization of PTWI through consumption of one cup daily per week of 200 mL of the product for a person weighing 70 kg (%)	
Pb	25 μg/kg	1750	0.0002 ± 0.0001 <LOD-0.0007	0.01	

LOD for Co = 0.0001 mg/200 mL; LOD for Ni = 0.00004 mg/200 mL; LOD for Pb = 0.0001 mg/200 mL

[a] American recommendations [44]

[b] Polish recommendations [43]

[c] In the form of vitamin B_{12}

There were also assessed levels of heavy metals such as Pb (Table 7) and Cd in the infusions, but the latter was under the limit of detection of the method applied. Thus, there could not have been estimated its provisional tolerable weekly intake (PTWI) realization. Former PTWI dose for Pb as recommended by the WHO/FAO [48] should not exceed 25 μg/kg, but it was withdrawn by the 73rd report of the Joint FAO/WHO Expert Committee of Food Additives [49]. It was found that it is not possible to establish a new dose PTWI that would be health protective. The PTWI's analyses were based on the earlier guidelines of WHO/FAO [48] for a person weighing 70 kg. The average Pb levels in 200 mL beverage amounted to 0.0002 mg. Therefore, the consumption of one cup daily per week of 200 mL tea results in the realization of PTWI for Pb in 0.01 %. It can be concluded that drinking black tea does not result in exceeding PTWIs; thus, it poses no health risk for human.

Conclusions

Although specified regulations concerning tea quality are not established in the European Union, it is important to control it as this beverage is one of the most commonly consumed. Therefore, we determined the elemental composition of various black tea samples originating from China, India, Ceylon, and Kenya. We conclude that there is no significant health risk associated with consumption of the analyzed tea samples, but that tea can constitute a valuable source of manganese in the human diet. Based on the obtained elemental data, the percentage of leaching as well as daily intake realization were assessed. The highest level of RDAs' realization was noted for Mn (15 %) and Co (10 %). Verification of the interdependences between elemental composition and country of origin of samples was done by multivariate techniques such as factor analysis and cluster analysis. They allowed on differentiation of teas according to the country of origin, i.e., China, India, Ceylon, and Kenya. Moreover, they were found helpful in diversification of teas originating from various plantations within a single country. Thus, they proved to be good tools able to differentiate samples in view of their provenance as well as plantation within the common region.

Compliance with Ethical Standards The manuscript does not contain clinical studies or patient data.

Conflict of Interest The authors declare that they have no conflict of interest.

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Recipient Justyna Brzezicha-Cirocka, MSc.

References

1. FAO (Food and Agriculture Organization) (2011) FAOSTAT. http://faostat3.fao.org/download/FB/CC/E. Accessed: May 2016
2. FAO (Food and Agriculture Organization) (2012) FAOSTAT. http://faostat3.fao.org/download/FB/CC/E. Accessed: May 2016
3. Wojciechowska-Mazurek M, Starska K, Mania M, Rebeniak M, Karłowski K (2010) Pierwiastki szkodliwe dla zdrowia w herbacie – ocena zagrożenia dla zdrowia. Bromat Chem Toksykol 43:233–239 in Polish
4. Barone G, Giacominelli-Stuffler R, Storelli MM (2016) Evaluation of trace metal and polychlorinated biphenyl levels in tea brands of different origin commercialized in Italy. Food Chem Toxicol 87:113–119
5. Ma G, Zhang Y, Zhang J, Wang G, Chen L, Zhang M, Liu T, Liu X, Lu C (2016) Determining the geographical origin of Chinese green tea by linear discriminant analysis of trace metals and rare earth elements: taking *Dongting Biluochun* as an example. Food Control 59:714–720
6. Fernández PL, Pablos F, Martin MJ, González AG (2002) Multi-element analysis of tea beverages by inductively coupled plasma atomic emission spectrometry. Food Chem 76:483–489
7. Wang L, Wei K, Cheng H, He W, Li X, Gong W (2014) Geographical tracing of Xihu Longjing tea using high performance liquid chromatography. Food Chem 146:98–103
8. Wu QJ, Dong QH, Sun WJ, Huang Y, Wang QQ, Zhou WL (2014) Discrimination of Chinese teas with different fermentation degrees by stepwise linear discriminant analysis (S-LDA) of the chemical compounds. J Agr Food Chem 62:9336–9344
9. Lin J, Zhang P, Pan Z, Xu H, Luo Y, Wang X (2013) Discrimination of oolong tea (*Camellia sinensis*) varieties based on feature extraction and selection from aromatic profiles analysed by HS-SPME/GC-MS. Food Chem 141:259–265
10. Qin Z, Pang X, Chen D, Cheng H, Hu X, Wu J (2013) Evaluation of Chinese tea by the electronic nose and gas chromatography-mass spectrometry: correlation with sensory properties and classification according to grade level. Food Res Int 53:864–874
11. Ye N, Zhang L, Gu X (2012) Discrimination of green teas from different geographical origins by using HS-SPME/GC-MS and pattern recognition methods. Food Anal Method 5:856–860
12. Lee JE, Lee BJ, Chung JO, Hwang JA, Lee SJ, Lee CH (2010) Geographical and climatic dependencies of green tea (*Camellia sinensis*) metabolites: a H^1NMR based metabolomics study. J Agr Food Chem 58:10582–10589
13. Ren G, Wang S, Ning J, Xu R, Wang Y, Xing Z (2013) Quantitative analysis and geographical traceability of black tea using fourier transform near-infrared spectroscopy (FT-NIRS. Food Res Int 53:822–826
14. Fernández-Cáceres PL, Martín MJ, Pablos F, Gonzalez AG (2001) Differentiation of tea (*Camellia sinensis*) varieties and their geographical origin according to their metal content. J Agr Food Chem 49:4775–4779
15. Herrador MA, González AG (2001) Pattern recognition procedures for differentiation of green, black and oolong teas according to their metal content from inductively coupled plasma atomic emission spectrometry. Talanta 53:1249–1257
16. Moreda-Piñeiro A, Fisher A, Hill SJ (2003) The classification of tea according to region of origin using pattern recognition techniques and trace metal data. J Food Compos Anal 16:195–211
17. Szymczycha-Madeja A, Welna M, Pohl P (2012) Elemental analysis of teas and their infusions by spectrometric methods. Trends Anal Chem 35:165–181
18. Paz-Rodríguez B, Domínguez-González MR, Aboal-Somoza M, Bermejo-Barrera P (2015) Application of high resolution-continuum source flame atomic absorption spectrometry (HR-CS FAAS): determination of trace elements in tea and tisanes. Food Chem 170:492–500
19. Brzezicha-Cirocka J, Grembecka M, Szefer P (2016) Monitoring of essential and heavy metals in green tea from different geographical origins. Environ Monit Assess 188:1–11
20. Brzezicha-Cirocka J, Grembecka M, Szefer P (2016) Analytical assessment of bio- and toxic elements distribution in Pu-erh and fruit teas in view of chemometric approach. Biol Trace Elem Res. doi:10.1007/s12011-016-0669-4
21. Konieczka P, Namieśnik J (2009) Quality assurance and quality control in the analytical chemical laboratory: a practical approach. CRC Press - Taylor & Francis Group, Boca
22. Szefer P (2007) Chemometric techniques in analytical evaluation of food quality. In: Szefer P, Nriagu JO (eds) Mineral components in foods. CRC Press - Taylor & Francis Group, Boca Raton
23. Dambiec M, Polechonska L, Klink A (2013) Levels of essentials and non-essential elements in black teas commercialized in Poland and their transfer to tea infusion. J Food Compos Anal 31:62–66

24. Soomro MT, Zahir E, Mohiuddin S, Khan AN, Naqvi II (2008) Quantitative assessment of metals in local brands of tea in Pakistan. Pak J Biol Sci 11:285–289
25. Yemane M, Chandravanshi BS, Wondimu T (2008) Levels of essential and non-essential metals in leaves of the tea plant (*Camellia sinensis* L.) and soil of Wushwush farms, Ethiopia. Food Chem 107:1236–1243
26. Gebretsadik DW, Chandravanshi BS (2010) Levels of metals in commercially available Ethiopian black teas and their infusions. Bull Chem Soc Ethiop 24:339–349
27. Malik J, Szakova J, Drabek O, Balik J, Kokoska L (2008) Determination of certain micro and macroelements in plant stimulants and their infusions. Food Chem 111:520–525
28. Gallaher RN, Gallaher K, Marshall AJ, Marshall AC (2006) Mineral analysis of ten types of commercially available tea. J Food Compos Anal 19:53–57
29. Shaltout AA, Abd-Elkader OH (2016) Levels of trace elements in black teas commercialized in Saudi Arabia using inductively coupled plasma mass spectrometry. Biol Trace Elem Res. doi:10.1007/s12011-016-0728-x
30. Maupenzi JP, Li L, Ge J, Varenyam A, Habiyaremye G, Theoneste N, Emmanuel K (2011) Assessment of soil degradation and chemical compositions in Rwandan tea-growing areas. Geosci Front 2: 599–607
31. Al-Oud SS (2003) Heavy metal contents in tea and herb leaves, Pakistan. J Biol Sci 6:208–212
32. McKenzie JS, Jurado JM, Pablos F (2010) Characterisation of tea leaves according to their total mineral content by means of probabilistic neural networks. Food Chem 123:859–864
33. Milani RF, Morgano MA, Cadore S (2016) Trace elements in *Camelia sinensis* marketed in southern Brazil: extraction from tea leaves to beverages and dietary exposure. LWT-Food Sci Technol 68:491–498
34. Wong MH, Hang ZQ, Wong JWC, Lan CY (1998) Trace metal contents (Al, Cu and Zn) of tea: tea and soil from two tea plantations, and tea products from different provinces of China. Environ Geochem Health 20:87–94
35. Han WY, Zhao FJ, Shi YZ, Ma LF, Ruan JY (2006) Scale and causes of lead contamination in Chinese tea. Environ Pollut 139:125–132
36. Seenivasan S, Manikandan N, Muraleedharan NN, Selvasundaram R (2008) Heavy metal content of black teas from south India. Food Control 19:746–749
37. Görür FK, Keser R, Akçay N, Dizman S, Okumuşoğlu NT (2011) Radionuclides and heavy metals concentrations in Turkish market tea. Food Control 22:2065–2070
38. Franklin RE, Duis L, Brown R, Kemp T (2005) Trace element content of selected fertilizers and micronutrient source materials. Commun Soil Sci Plan 36:1591–1609
39. De Mee s C, Eduljee GH, Hutton M (2002) Assessment and management of risk arising from exposure to cadmium in fertilisers. Sci Total Environ 291:167–187
40. Lv HP, Zhang YJ, Lin Z, Liang YR (2013) Processing and chemical constituents of Pu-erh tea: a review. Food Res Intern 53:608–618
41. Sarma H, Islam NF, Borgohain P, Sarma A, Prasad MNV (2016) Localization of polycyclic aromatic hydrocarbons and heavy metals in surface soil of Asia's oldest oil and gas drilling site in Assam, north-east India: implications for the bio-economy. Emerging Contaminants. doi:10.1016/j.emcon.2016.05.004
42. Hua D, Ming-Shun L, Yu-Chan Z (2012) Soil metal contamination and fractionation of tea plantations: case studies in a normal tea garden and in a restored mineland tea stand. Pol J Environ Stud 21:1223–1228
43. Jarosz M (2012) Normy żywienia dla populacji polskiej – nowelizacja. IŻŻ, Warsaw in Polish
44. American recommendations (2011) Food and Nutrition Board, Institute of Medicine, National Academies Press. URL https://fnic.nal.usda.gov/sites/fnic.nal.usda.gov/files/uploads/recommended_intakes_individuals.pdf. Accessed May 2016
45. Powell JJ, Trevor JB, Thompson RPH (1998) *In vitro* mineral availability from digested tea: a rich dietary source of manganese. Analyst 123:1721–1724
46. WHO (2011a) Manganese in drinking-water, background document for preparation of WHO guidelines for drinking water quality. World Health Organization, Geneva. URL http://www.who.int/ipcs/publications/cicad/cicad69%20.pdf. Accessed 3 Apr 2016
47. Kabata-Pendias A, Szteke B (2015) Trace elements in abiotic and biotic environments. CRC Press - Taylor & Francis Group, Boca Raton
48. WHO (2010) Lead in drinking-water, background document for preparation of WHO Guidelines for drinking-water quality. World Health Organization, Geneva. URL http://www.who.int/water_sanitation_health/dwq/chemicals/lead.pdf. Accessed 3 Apr 2016
49. WHO (2011b) Seventy-third report of the Joint FAO/WHO Expert Committee on Food Additives. World Health Organization, Geneva. URL http://whqlibdoc.who.int/trs/WHO_TRS_960_eng.pdf. Accessed 3 Apr 2016