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Monitoring content of cadmium, calcium, copper, iron, lead, magnesium and manganese in tea leaves by electrothermal and flame atomizer atomic absorption spectrometry

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Abstract: Due to the simplicity of tea preparation (pouring hot water onto different dried herbs) and its high popularity as a beverage, monitoring and developing a screening methodology for detecting the metal content is very important. The concentrations of Cd, Ca, Cu, Fe, Pb, Mg and Mn in 11 different samples of sage (Salvia officinalis L.), linden (Tilia L.) and chamomile (Matricaria chamomilla L.) purchased at local herbal pharmacy were determined using electrothermal atomizer atomic absorption spectrometry (ETAAS) and flame atomizer atomic absorption spectrometry (FAAS). The concentrations determined were: Cd (0.012 -0.470 mg kg⁻¹), Ca (5209 - 16340 mg kg⁻¹), Cu (22.01 -33.05 mg kg⁻¹), Fe (114.2 - 440.3 mg kg⁻¹), Pb (0.545 - 2.538 mg kg^{-1}), Mg ($2649 - 4325 \text{ mg kg}^{-1}$) and Mn (34.00 -189.6 mg kg⁻¹). Principal Component Analysis (PCA) was applied to identify factors (soil and climate) influencing the content of the measured elements in herbal samples. The proposed methodology developed in this work was successfully applied to the detection of metals in herbal

samples. The analysis showed that the content of toxic metals in herbal teas was below the maximum dose recommended by the World Health Organization (WHO).

Keywords: trace elements, chemometrics, tea, ETAAS, FAAS, PCA

1 Introduction

Tea has been part of our lives for a long time. The first written records reporting tea consumption originated in ancient China five thousand years ago. Tea drinking has not slowed down since those ancient times: according to data provided by the Croatian Bureau of Statistics, the annual consumption of herbal teas and tea products in Croatia has increased by 12% over the past several years [1]. Recently, however, tea drinkers have become aware of contaminants, especially heavy metals, that pose potential health hazards. It is very important to monitor the metal concentrations in daily food and drink content and intake. This is especially critical among common beverages like teas, since the metals extracted from the leaves, where plants tend to store cadmium and other metals extracted from the soil, during tea preparation can have beneficial or harmful effect on human health [2]. Due to these factors, the analysis and detection of heavy metals in herbal teas and tea related products has recently attracted significant interest.

In Croatia, the main herbs used for tea brewing are chamomile, linden, sage, and mint, while fruits such as rose hip, blueberry, and peach are also used. This work reports a continuation of our earlier work in the analysis of trace metal content in widely used dietary products, especially herbal teas and olive oil, from Croatian markets [2,3]. It is very difficult to find published reports dedicated to minerals and trace metals in chamomile, linden and sage [4]. The vast majority of available manuscripts, e.g.

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[5-7], focus on the most popular teas consumed in the world - black and green tea.

Minerals and trace elements have a number of key roles in the human body, for example, calcium (Ca) is important for the growth and health of bones and teeth [8], iron (Fe) is essential for oxygen transport [9], and zinc (Zn) and copper (Cu) are essential to the function of many key enzymes [10,11]. Metals can have toxic effects as well, however. Cadmium (Cd), in particular, has been recognized as a human [12] and multi-tissue animal carcinogen [13]. Over half of cadmium in soils arises during the distribution of phosphatic fertilizers [14-16]. Cd is also well known for its phytotoxicity [17], which manifests as an inhibition of plant growth [18], nitrate assimilation [19] and photosynthesis [20,21], as well as disturbances in plant ion [22] and water balances.

Some metals are considered "biogenic," as they exist in significant concentrations in living tissues. These metals can be toxic to humans, but levels required to reach toxicity tend to be relatively high. Two such metals are calcium (Ca) and magnesium (Mg). Calcium has a number of important physiological functions such as maintenance of the rhythm of cardiac muscle cells and the reduction of excitability of both nerves and muscles [8]. Hypercalcemia (when the Ca concentration in an organism is above 2.6 mM) could be related to the development of myeloma, hyperparathyroidism and vitamin D intoxication [23]. On the other hand, hypocalcemia, which is a risk factor for osteoporosis, is typically caused by hypoparathyroidism, vitamin D deficiency, or renal failure [23]. Studies have shown that magnesium (Mg) is a vital mineral for humans. Mg is needed for normal functioning of the heart, muscles and nerves. It also participates in the activation of more than 300 enzymes that ensure the smooth operation of many metabolic processes. Since Mg cannot be synthesized in the human body, it must be taken by the ingestion of nutrients [33,34].

Other metals are not thought to be required at any concentration by most organisms. These metals are considered "toxic" because they lead to harmful effects even at very low concentrations. Two such metals are cadmium (Cd) and lead (Pb). Lead (Pb) was identified very early in human history and is very well documented as one of the most common contaminants in the environment [29]. Humans are usually exposed to lead through occupational and environmental sources; for instance, if water contaminated with Pb was used for herbal tea brewing, it could lead to Pb poisoning [30,31].

A third classification of metals is "biogenic/toxic." These metals, including copper (Cu), iron (Fe), and manganese (Mn), are essential for life at trace levels but

become toxic at fairly low concentrations. Copper (Cu) is one of the micronutrients found in plants and animals, but it becomes toxic in adult humans above a daily intake of 2 mg [24]. Copper can be found on the crops due to the usage of various cupreous compounds in agro-technical treatments [24,25]. Due to the fact that Cu can be biogenic and toxic, Cu content in our surroundings, such as in our water, food, and beverages, has to be traced and controlled consistently [24-28]. Iron (Fe) is an essential nutrient for all forms of life [9]. Fe is a cofactor for many enzymes and it is essential for oxygen transport (in hemoglobin) and electron transfer [9]. In humans, the daily requirement for Fe varies between 8-18 mg, but it can be toxic in excess concentrations because of its pro-oxidant activity [22].

Due to the significance of minerals and trace metals in human health, the World Health Organization (WHO) recommends a dietary allowance (RDA). Amongst all metals covered in the present report, WHO classifies Ca and Mg as minerals and Cu, Fe and Mn as trace metals. The RDA for these metals are as follows: Ca, 1.3 g; Mg, 0.42 g; Cu, 0.9 mg; Fe, 18 mg; and Mn, 2.3 mg. In this manuscript, we quantified metal content in tea samples from a local herbal pharmacy. The tea samples were carefully selected as they represented the best-selling herbal teas in Croatian markets. The aim of this study was to identify differences in the metal content of the samples, which depended primarily on herb variety but also on treatments applied during the sample production.

2 Materials and Methods

Measurements of Cd and Pb were carried out using a Model AAS vario 6 ETAAS atomic absorption spectrometer (Analytik Jena AG, Jena, Germany, 2001) equipped with a transversely heated graphite atomizer with autosampler (Model MPE 50), a deuterium background correction system and a hollow cathode lamp. For Cd, the lamp operated at 3 mA (wavelength 228.8 nm) and for Pb, it operated at 3 mA (wavelength 283.3 nm). Pyrolytic coated graphite tubes with PIN-platform (Analytik Jena, Part No. 407-A81.025) were used during the analytical determination (Analytik Jena AG, 2001). The injection volume was 20 μ L and integrated absorbance (peak area) was used for signal evaluation.

Ca, Cu, Fe, Mg and Mn measurements were carried out using a Model AAS vario 6 FAAS atomic absorption spectrometer equipped with deuterium background correction system and a hollow cathode lamp for calcium, copper, iron and manganese (wavelength 422.7 nm for Ca; 324.8 for Cu nm; 248.3 nm for Fe and 279.5 nm for Mn). Concentrations of Cu, Fe and Mn were determined by Flame AAS with a C₂H₂/air burner, while Ca and Mg were determined with a C₂H₂/N₂O₂ burner (Analytik Jena AG, 2001). Argon (5N purity) was used as the purge gas at 300 mL min⁻¹, except in the atomization stage (gas stop). Acetylene, air and nitrogen (II) oxide were mixed on the burner and used for atomization by the flame. All of the gases used were 4N purity unless otherwise noted. The samples were weighed using a Mettler AX 205 (Mettler Toledo, Columbus, Ohio, United States of America) electronic balance. Analytical measurements were based on the absorbance peak area as described [36].

The basic instrumental and experimental conditions for ETAAS determinations are shown in Table 1. Hollow cathode lamps (HCL) were used for all measurements. All ETAAS and FAAS measurements were carried out in triplicate and all of the results are presented as the arithmetic mean of measured values according to the standard method HRN EN 14084:2005, which corresponds to the British Industrial Standard EN 14084:2003. Recoveries were found to be within the range of 95 to 102 %.

Tea leaf samples were digested in a closed microwave system, CEM Model Mars 5 (CEM Corporation, United States of America), which is a microwave oven equipped with internal pressure and temperature control systems. This oven has a variable power range (up to 630 W) adjustable in 1 % increments and a programmable timer. Lined Teflon® vessels with a volume of 100 mL and a pressurerelief valve were used. The temperature and pressure in the vessel during wet digestion were between 210°C and 240°C and 0.70 MPa and 1.0 MPa, respectively. The first cycle of microwave digestion lasted for 25 minutes and the second one 15 minutes. After a cooling down period, samples were transferred into 50 mL volumetric flasks and diluted with supra pure water (with a conductivity of 0.04 uS cm⁻¹) produced using the Millipore Simplicity system (Millipore, United States of America). Table 1 shows the physical properties of ETAAS employed in this work, and Table 2 displays the linear range, limit of detection (LOD), limit of quantification (LOQ) and relative standard deviation (RSD) for Cd, Ca, Cu, Fe, Pb, Mg and Mn standard solutions used for preparing calibration curves.

Samples of commercially available teas were purchased from a local pharmacy (samples A1 to A11, Table 3). A mixture of plant leaves and twigs formed the samples. Depending on the plant material structure, the sample mass ranged from 0.30 to 0.45 g. When we made comparisons among collected results, all concentrations were converted to mg kg^{-1} of dried herbal material as shown in Tables 4 to 6.

Table 1: Basic instrumental parameters of ETAAS for the determination of Cd and Pb in tea samples.

	Cd	Pb	
Wavelength, nm	228.8	283.3	
Slit/nm	0.5R	0.5R	
Purge gas	Ar	Ar	
Drying temperature, °C	120	120	
Ramp, hold, s	72/50	72/50	
Ashing temperature, °C	900	1200	
Ramp, hold, s	13/10	4/4	
Atomization, °C	1300	2050	
Ramp, hold, s	3.3/3	5/4	
Clean-up, °C	2300	2300	
Hold, s	4	4	
L'vov platform	Yes	Yes	
Integration time, s	4	4	
Injected sample volume, µL	20	20	
Modifier volume, μL	5	3	

Each sample was put in Teflon® vessels with 4 mL of nitric acid and 2 mL of hydrogen peroxide for microwave digestion.

Almost all Croatian industrial growth of chamomile plants is based in Slavonia. Slavonia is part of Croatia with abundant plains used for agricultural production. Due to the use of artificial fertilizers, soil in Slavonia is laden with different heavy metals such as Cd and Cu. Wild plants of sage and linden are dispersed throughout Croatia where it can be easily harvested and taken to factories for tea production.

All solutions were prepared by dissolving the chemicals in supra pure water. Hydrogen peroxide, s.p. (supra pure) and nitric acid, s.p., were obtained from Merck (Darmstadt, Germany). All standard solutions had metals in the form of metal nitrate in nitric acid solution with concentrations of 1000 ± 2 mg L⁻¹ (Merck, Darmstadt, Germany). All matrix modifiers employed, such as palladium nitrate, p.a. (pro analysis) and magnesium nitrate, p.a., were also obtained from Merck. Pyrolysis and atomization curves were established in the presence of a chemical modifier: 0.1% Pd(NO_3)₂ + 0.05% Mg(NO_3)₂•6H₂O. The modifier was prepared from a commercially available stock solution (Merck): Art.1.07289 palladium matrix for graphite furnace AAS and Art.1.05855 magnesium nitrate hexahydrate. The final volume of modifier added was 5 μ L.

Statistical data analyses were made using the R program ver. 2.9.2 [35]. Principal component analysis (PCA) was applied to analyze the significance of each element in the data structure.

Teas purchased at drugstores were dried and produced in factories in different ways depending on the plant source material.

Table 2: Linear range, LOD, LOQ and RSD for Cd, Ca, Cu, Fe, Pb, Mg and Mn in FAAS analyses.

Parameters	Cd	Ca	Cu	Fe	Pb	Mg	Mn
Linear range/	0.20-1.00	500-4000	100-500	100-1000	10-50	100-1000	100-1000
μg L ⁻¹							
LOD/µg L ⁻¹	0.08	26	28	26	0.07	18	20
LOQ/µg L ⁻¹	0.26	86	94	88	0.24	62	66
RSD/%	2.00	1.60	1.89	0.56	2.30	1.53	2.25

LOD - limit of detection

LOO - limit of quantification

RSD - relative standard deviation

Table 3: Descriptions of investigated herbal tea samples.

Sample	Туре	Herb latin name	Origin	Trademark
1	Tea bags	Salvia officinalis L.	Croatia/wild herb	Holyplant
2	Tea bags	Salvia officinalis L.	Croatia/wild herb	Biofarm
3	Tea bags	Tilia L.	Croatia/wild herb	Franck
4	Tea bags	Tilia L.	Croatia/wild herb	Podravka
5	Tea bags	Tilia L.	Croatia/wild herb	Travar MB
6	Tea bags	Salvia officinalis L.	Croatia/wild herb	Travar MB
7	Tea bags	Matricaria chamomilla L.	Croatia/industrial growth	Pharmacy of SD County
8	Bulk	Matricaria chamomilla L.	Croatia/industrial growth	Palak
9	Bulk	Matricaria chamomilla L.	Croatia/industrial growth	Suban
10	Bulk	Matricaria chamomilla L.	Croatia/industrial growth	Home Production
11	Tea bags	Matricaria chamomilla L.	Croatia/industrial growth	Agristar

3 Results and Discussion

The detection limits for Cd and Pb analyzed by the ETAAS technique were found to be 1 µg L⁻¹. Matrix effects and interferences by other metals were minimized with the use of a graphite platform and different modifiers (0.1 % $Mg(NO_3) + 0.05 \% Pd(NO_3)$. The detection limits using the FAAS technique were: Ca, 100 μg L⁻¹; Mg, 200 μg L⁻¹; Cu, $50 \mu g L^{-1}$; Fe, $50 \mu g L^{-1}$ and Mn, $10 \mu g L^{-1}$. Table 4 shows the calculated and the measured concentrations of each tested metal in every sample as well as the means, standard deviations (SD), minima, maxima, and medians.

Table 4 illustrates that different herbal tea samples contain significantly different concentrations of heavy In analyzed samples, the metals that were present at the highest concentration were Ca and Mg, which represented 1.00 \pm 0.41 g kg¹ and 3.56 \pm 0.51 g kg¹, respectively. Mn, Fe, and Cu were present at lower concentrations (121.23 \pm 56.280 mg kg⁻¹, 207.95 \pm 124.05 mg kg $^{-1}$ and 26.154 \pm 3.696 mg kg $^{-1}$, respectively). However, the lowest contents were recorded for Pb and Cd (1.0386 ± $0.5736 \text{ mg kg}^{-1}$ and $0.1731 \pm 0.1521 \text{ mg kg}^{-1}$, respectively).

We performed PCA to identify factors that might influence the content of elements present in tea samples.

Factor 1 was represented by variables Fe, Cu, Ca, and Pb, while factor 2 was represented by Mg.

A highly negative contribution to factor 1 was found for all four samples of chamomile, while two samples of sage and one of linden had a strongly positive contribution. Also, a strong positive contribution to factor 2 was detected for all three samples of sage, while a strongly negative contribution was found for all samples of linden and two samples of chamomile. The projection of the tea samples on a factor-plane is presented in Figure 1, and as can be seen, three tea varieties were well divided from each other. The samples of sage were grouped in factor 1, the samples of linden in factor 2, while samples of chamomile negatively influenced factor 1. The presented results indicate and confirm the influence of the plant species on its metal content.

Other possible factors that might cause grouping of variables could not be detected, probably because the number of analyzed variables was relatively small. Since chamomile is cultivated, while sage and linden were collected from nature vegetation, it is also possible that principal component (PC) 1 was caused mainly by geogenic processes while factor 2 was caused by environmental processes specific to sage plants.

Table 4: Elemental content in analyzed tea samples expressed as mass of metal per kg of dry input sample (mean of three measurements).

Sample	Concentration							
	Cd/ µg kg ⁻¹	Ca/g kg ⁻¹	Cu/ mg kg ⁻¹	Fe/ mg kg ⁻¹	Pb/ µg kg⁻¹	Mn/ mg kg ⁻¹	Mg/ g kg ⁻¹	
1	18.7	11.7	27.1	127	811	37.0	4.18	
2	23.1	13.6	26.9	365	1618	54.1	3.80	
3	55.7	16.3	30.6	440	2538	115	3.00	
4	87.7	14.8	33.1	387	845	138	3.26	
5	126.2	12.1	29.9	125	1173	190	2.50	
6	11.9	11.1	22.2	147	765	34.0	3.76	
7	292.2	5.30	24.6	139	768	164	3.45	
8	279.3	6.17	23.1	136	712	128	4.00	
9	226.2	5.21	23.6	131	893	126	3.52	
10	469.5	8.34	24.7	177	757	167	4.33	
11	313.7	5.84	22.0	114	545	180	3.23	
Mean	173.1	10.0	26.2	208	1039	121	3.56	
SD*	152.1	4.05	3.70	124	574	56.3	0.51	
Minimum	11.9	5.21	22.0	114	545	34.0	2.65	
Maximum	469.5	16.3	33.1	440	2538	190	4.33	
Median	126.2	11.1	24.7	139	811	1280	3.52	

Table 5: Factor loadings for metal content in tea samples of sage, linden and chamomile (n = 7).

	Rotated component matrix			
Component matrix	PC1	PC2		
Fe	0.86	-0.07		
Cu	0.82	-0.34		
Mn	-0.34	-0.93		
Ca	0.95	0.09		
Mg	-0.41	0.73		
Cd	-0.76	-0.44		
Pb	0.82	-0.12		
Eigenvalue	3.8	1.7		
Proportion of total variance	0.55	0.25		

In order to explain the grouping of variables the PCA should be conducted on more variables per tea sample. Analysis using Cd, Ca, Cu, Fe, Pb, Mg and Mn was able to distinguish the botanical origin of the tea, and effect that is illustrated in Table 5.

Finally, PCA analysis was followed by a t-test in order to compare the content of metals in sage, linden and chamomile leaves. The average concentrations of Ca, Cu, Mn and Mg in sage, linden and chamomile were compared

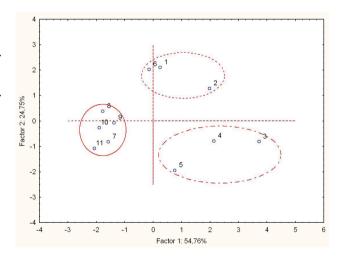


Figure 1: PCA diagram of interrelations of 11 teas based on Ca, Cd, Cu, Fe, Mg, Mn and Pb content.

by t-test for independent samples. Since the test of variance homogeneity failed for Cd, Fe and Pb, the mean content of those metals was performed by nonparametric statistics for independent samples using Kolmogorov-Smirnov test to compare mean values. The results of these statistical analyses are displayed in Table 6.

Among tea samples, there is a significant difference in the content of Cd, Ca, Cu, Mn and Mg. Table 6 illustrates that the content of Fe and Pb were found to be similar among analyzed herbal samples. The variability of some of the content of some of the metals could be attributed to the high variability among the herbs and country of origin.

The results reported in this manuscript were compared with similar investigations reported in Croatia [2], Poland [4] and Serbia [37,38] as illustrated in Table 7. We emphasize that the results reported from Serbian teas were related to an area which is approximately 600 km away and where the climate is humid subtropical as compared to the continental and Mediterranean climate in south Croatia. In addition, there were also differences in the soil composition: the Serbian soil was chernozem and dilluvial whereas the Croatian soil included "terra rosa" and chernozem. Polish herbs are grown in an area that is about 1200 km away from south Croatia, with climate and soil conditions similar to those in Serbia.

A comparison of the results displayed in Table 7 shows that the content of Ca and Mg in plants is very

Table 6: Results of statistical comparison among sage, linden and chamomile with respect to content of Cd, Ca, Cu, Fe, Pb, Mn and Mg, expressed in mg kg⁻¹. Means with the same letter are similar at P=0.05 level of significance.

	Sage	Linden	Chamomile
Cd*	0.020a	0.090ª	0.316 ^b
Ca	12123 ^b	14396 ^b	6170ª
Cu	25.40ª	31.19 ^b	23.59ª
Fe*	212.9 ^a	317.4ª	139.3ª
Pb*	5.92ª	12.65 ^a	0.735ª
Mn	41.71 ^a	147.7 ^b	153.1 ^b
Mg	3912 ^b	2969ª	3704 ^b

^{*}Nonparametric analyses for independent samples was performed; a, b - mean values were compared with the Kolmogorov-Smirnov test of significance.

similar to what was expected. This could be attributed to the importance of these elements in plant metabolism and generally for essential biological processes within the plant. We established that the content of Ca and Mg in a tea product can be used both for identification of the source plant as well as for an indicator of poor soil [2,4,37,38]. Although the amount of Ca and Mg in tea is related to the total Ca and Mg content in herbs, it is possible to use teas as dietary source of Ca and Mg [4,39].

The situation as above described for Ca and Mg was observed for Cu, Fe and Mn. Mn content in our investigation was found to be lower than in samples purchased at the supermarkets [2]. However, in all other cases, Mn content was found to be higher than those described in previous reports [37,38]. Practically all chamomile samples investigated had high Mn concentrations, which agrees with other comparable plants found in industrial production of teas and other pharmaceutical products. Since the Cu content found in all three plants had similar values, it is reasonable to conclude that Cu content is directly connected with its absorption, since the plantations for all tested plants were located in the same climate region and had the same soil type. In our earlier study [2] and in similar cases reported in the literature [37,38], the Cu content was found to be significantly lower (by a factor of 5-10). Statistical tests (t-test and Kolmogorov-Smirnov test) demonstrated that these results are not related to experimental error; therefore, this finding supports our hypothesis.

According to the results for Fe content displayed in Table 3, it is not possible to determine which plant preferentially accumulates Fe from the soil. The considerable differences in Fe content among the plants from the same species indicate that Fe content is probably more closely related to the production of the tea rather than a biological process. Cd and Pb contents are similar

Table 7: Comparison of the previously published results for Croatian, Polish and Serbian tea samples expressed as mg metal per kg of dry plant material (mean±SD).

Reference	Ca(%)	Cd	Cu	Fe	Mg(%)	Mn	Pb
[37]	0.90±0.40	N/A	10.94±0.60	405±3	0.53±0.02	111±2	N/A
[38]	1.51	0.26	11.37	117.5	0.49	64.1	3.45
[4]	a) 0.038±0.002	N/A	a) 3.22±0.06	a) 55.8±0.4	a) 0.020±0.001	a) 7.88±0.06	N/A
	b) 0.041±0.003		b) 1.75±0.02	b) 37.9±0.5	b) 0.042±0.002	b) 12.0±0.1	
	c) 0.018±0.002		c) 1.55±0.03	c) 16.8±0.3	c) 0.028±0.002	c) 13.8±0.1	
	d) 0.051±0.005		d) 1.67±0.04	d) 67.1±0.4	d) 0.032±0.002	d) 10.5±0.1	
[2] (marketplace)	N/A	0.170±0.174	4.69±0.84	134±80	0.54±0.516	158±231	1.46±0.237
[2] (supermarket)	N/A	0.252±0.050	3.08±1.10	451±437	0.0774±0.0622	66±26	0.522±0.590
This investigation	1.00±0.41	0.173±0.152	26.2±3.70	208±124	0.356±0.051	121±6.28	1.04±0.57

N/A - not available

a) Linden; b) mint; c) chamomile; d) sage

in our study and in the literature [2,37,39]. This fact is very important because it suggests that both Serbian and Croatian soils are not contaminated significantly with these toxic metals.

A comparison of the average daily dietary intake with the values of metal content in infusions of herbal tea grown in Croation soils [4] demonstrates that consumption of teas is not dangerous, as the content of toxic did not exceed 5% of total metal content in all cases, according to the literature [4,39].

4 Conclusion

Due to the potential toxicity or biogenic characteristics of heavy metals, it is critical to control the total metal content in foods and beverages consumed by humans. In this manuscript, we reported for the first time a significant correlation between metal amounts in different herbal teas, including sage, linden and chamomile that were grown in Croatia. The Ca, Cd, Cu, Mn and Pb contents correlate with plant type (with chamomile having the highest concentrations and sage having the lowest (Table 6)). Note that Cd, Cu, Mn and Pb are especially consistent, as their concentrations are practically the same in a given species of plant. The largest data dispersion was recorded for Fe content, which could be attributed to the processing of the herb during tea production.

Finally, we found that tea samples purchased at local drugstores had lower metal contents in comparison to those purchased at a local supermarket and in the marketplace. This discrepancy indicates the need for more rigorous controls of herbal products.

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