Original article

Trace heavy metal contents of some spices and herbal plants from western Anatolia, Turkey

Umit Divrikli,[1]* Nesrin Horzum,[1] Mustafa Soylak[2] & Latif Elci[1]

1 Department of Chemistry, Faculty of Art and Sciences, Pamukkale University, 20020, Denizli, Turkey
2 Department of Chemistry, Faculty of Art and Sciences, Erciyes University, 38039, Kayseri, Turkey

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Summary Trace metal levels in eleven different spice and herbal plant species from western Anatolia, Turkey were determined by atomic absorption spectrometry. The contents of trace metals in the herbal plant samples were found in the ranges: 3.8–35.4 $\mu g\ g^{-1}$ for copper, 0.2–2.7 $\mu g\ g^{-1}$ for cadmium, 0.1–2.8 $\mu g\ g^{-1}$ for lead, 1.4–11.3 $\mu g\ g^{-1}$ for nickel, 0.1–9.7 $\mu g\ g^{-1}$ for chromium, 30.0–945.3 $\mu g\ g^{-1}$ for iron, 7.9–152.5 $\mu g\ g^{-1}$ for manganese and 5.2–83.7 $\mu g\ g^{-1}$ for zinc. Results obtained are in agreement with data reported in the literature.

Keywords Atomic absorption spectrometry, herbal plants, spices, traces heavy metals.

Introduction

Heavy metals have important positive and negative roles in human life (Adriano, 1984; Slaveska *et al*., 1998; Divrikli *et al*., 2003; Dundar & Saglam, 2004; Colak *et al*., 2005; Oktem *et al*., 2005). Some of the heavy metals are considered essential including iron, zinc and copper. Some metal ions like cadmium, lead and mercury have toxic roles in biochemical reactions on our body. There is a strong link between micronutrient nutrition of plants, animals and humans and the uptake and impact of contaminants in these organisms (De Leonardis *et al*., 2000; Yuzbasi *et al*., 2003; Baslar *et al*., 2005; Yaman *et al*., 2005). The content of essential elements in plants is conditional, the content being affected by the characteristics of a soil and the ability of plants to selectively accumulate some metals. Additional sources of heavy metals for plants are: rainfall, traffic density, use of oil or fossil fuels for heating, atmospheric dusts, plant protection agents, and fertilizers, which could be adsorbed through the leaf blades (Kovacheva *et al*., 2000; Lozak *et al*., 2002; Atrouse *et al*., 2004). In addition, they could be contaminated from various species including trace metals as farmers wash them with waste water before bringing them to market.

Spices and herbal plants contain heavy metal ions over a wide range of concentrations. As each element has one or more specific structural or functional roles in the plant, in the absence of that element, the plant will be expected to exhibit certain morphological or biochemical symptoms of that deficiency (Hopkins, 1995; Chizzola *et al*., 2003; Ozcan, 2004; Lemos *et al*., 2005; Ozcan *et al*., 2005; Soylak *et al*., 2005). Some of these elements are toxic to humans even at considerably low concentrations. Especially, toxic trace heavy metals like cadmium and lead are known to pose a variety of health risks such as cancer, mutations or miscarriages (Weigert, 1991). People in western Anatolia region of Turkey consume large amounts of spices and herbal plants for various purposes, especially for healthy life. Therefore, the metal toxicity has attracted concern over safety of spices and herbal plants consumption in western Anatolia, Turkey.

In the present work, the concentrations of copper, iron, nickel, cadmium, manganese, lead, chromium and zinc in the spice and herbal plant samples purchased from farms around western Anatolia region of Turkey were investigated by flame atomic absorption spectrometry.

Materials and methods

Apparatus

The instrumental detection system used was a Perkin Elmer AAnalyst 700 model atomic absorption spectrometer (Norwalk, CT, USA). The operating parameters for working elements were set as recommended by the manufacturer. The parameters are given in Table 1. Reagent blank determinations were used to correct the instrument readings. Calibration standard solutions were prepared from stock metal standard solutions. The concentrations of analytes were obtained directly

*Correspondent: Fax: +90 258 2125546;
e-mail: udivrikli@pamukkale.edu.tr

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Table 1 Instrument settings

Elements	Parameters		
	Wavelength (nm)	Slit width (nm)	Lamp current (mA)
Cu	324.8	0.7	30
Fe	248.3	0.2	25
Ni	232.0	0.2	30
Cd	228.8	0.7	4
Mn	279.5	0.2	25
Pb	283.3	0.7	10
Cr	357.9	0.7	30
Zn	213.9	0.7	30

from calibration graphs after correction of the absorbance for the signal from an appropriate reagent blank.

Reagents

All chemicals were of reagent-grade and all solutions were prepared in distilled–deionised water. All the plastic and glassware were cleaned by soaking in dilute HNO_3 (1 + 9) and were rinsed with distilled water prior to use. The element standard solutions used for calibration were prepared by diluting stock solutions of 1000 mg L^{-1} of each element supplied from Sigma, St Louis.

Sampling and sample treatment

The spice and herbal plant samples were collected from western Anatolia, Turkey according to sampling strategy in Divrikli *et al.* (2003) and Kovacheva *et al.* (2000). Fifty farmers visited for sample collection. The spice and herbal plant samples purchased from farmers over a period from June to October 2003. The number for each of the plants was four and 2 kg of each samples were collected. The samples were stored in polythene bags until analysis.

The samples are rosemary (*Rosmarinus officinalis* L.), basil (*Ocimum basilicum*), laurel (*Laurus nobilis*), chard (*Beta vulgaris* L. var. cicla), marjoram (*Origanum majorana* L.), sheep sorrel (*Rumex acetosella*), centaury (*Centaurium erythraea*), yarrow (*Achillea millefolium*), marshmallow (*Allthae officinalis*), lavender (*Lavandula angustifolia*) and mullein (*Verbascum* spp.).

The samples were divided from root, leaf and flowers and scrubbed. The analysed parts of the investigated plants given in Tables 2 and 3 were especially selected according to their consumption of the people from western Anatolia. The samples were then washed and cut to simulate the human intake conditions better. They were dried at 25 °C for 3 days on a sheet of paper to eliminate excess moisture. The samples were oven-dried at 80 °C for 12 h and ground in an agate mortar until they could pass a 60 mesh sieve. They were then stored in a clean, dry and stoppered glass container.

For the analysis, the samples were weighted (1.0 g) and dissolved with 8 mL of the mixture of acids HNO_3–$HClO_4$–H_2O_2 (2:1:1). The samples were heated at 170 °C for 3 h, cooled down, and then 2 mL of H_2SO_4 and 8 mL of acid mixture were added. These solutions were centrifuged at 490 ***g*** for 5 min. The solutions were filled up to 25 mL with 1 M HNO_3 (Divrikli *et al.*, 2003). The metal contents of this solution were determined by atomic absorption spectrometry.

Results and discussion

A recovery test of the total analytical procedure was performed for some of the investigated metals in selected samples by spiking analysed samples with aliquots of metal standards and then reanalysing them. Acceptable recoveries (> 95%) were obtained for the analyte ions. The contamination was not a problem in the digestion procedure, as the level of the analyte ions in the blank digest were close to their detection limits. The relative standard deviations (RSD) were calculated from pooled

Table 2 The levels of metal ions in the leaves, flower and roots of spices and herbal plants

Element	Rosemary	Basil	Laurel		Marjoram		Marshmallow	Chard	
	Leaf	Leaf	Leaf	Root	Leaf	Root	Flower	Leaf	Root
Cu	9.2 ± 0.1*	20.1 ± 0.7	3.8 ± 0.1	9.5 ± 0.2	11.3 ± 0.1	7.4 ± 0.2	6.1 ± 0.5	10.1 ± 0.4	5.9 ± 0.4
Cd	ND†	1.7 ± 0.1	2.7 ± 0.2	0.8 ± 0.07	0.5 ± 0.04	ND	1.8 ± 0.2	2.4 ± 0.1	1.5 ± 0.3
Pb	ND	ND	0.2 ± 0.1	0.1 ± 0.03	ND	ND	ND	0.2 ± 0.04	2.8 ± 0.6
Ni	4.8 ± 0.1	6.7 ± 0.1	11.3 ± 0.8	4.9 ± 0.1	3.2 ± 0.3	2.9 ± 0.7	10.9 ± 0.5	9.4 ± 0.2	8.8 ± 1.1
Cr	6.0 ± 0.1	5.1 ± 0.1	2.7 ± 0.3	4.0 ± 0.1	9.7 ± 0.3	3.3 ± 0.2	4.1 ± 0.1	2.2 ± 0.2	2.5 ± 0.1
Fe	398.7 ± 3.2	945.3 ± 2.1	341.6 ± 16.9	194.0 ± 4.3	231.4 ± 1.5	137.8 ± 1.1	606.6 ± 10.2	335.6 ± 47.7	251.9 ± 9.5
Mn	34.2 ± 0.6	30.1 ± 0.3	152.5 ± 7.5	83.4 ± 1.2	35.4 ± 0.2	8.8 ± 0.4	33.3 ± 0.3	53.7 ± 8.2	22.1 ± 0.9
Zn	22.6 ± 0.3	11.2 ± 0.7	20.1 ± 0.1	5.2 ± 0.3	83.7 ± 1.0	21.5 ± 0.3	19.8 ± 0.2	19.8 ± 0.2	19.1 ± 0.2

*Expressed in μg g^{-1}, mean ± SD.
ND, not determined.

Table 3 The levels of metal ions in the leaves, flower and roots of spices and herbal plants

	Sheep sorrel		Mullein		Lavender		Yarrow		Centaury	
Element	**Leaf**	**Root**	**Leaf**	**Flower**	**Leaf**	**Flower**	**Flower**	**Root**	**Flower**	**Root**
Cu	20.1 ± 0.3*	11.5 ± 1.2	35.4 ± 0.9	18.6 ± 1.2	11.3 ± 0.1	15.5 ± 0.4	17.6 ± 0.6	9.5 ± 0.3	11.3 ± 0.1	9.4 ± 0.2
Cd	1.6 ± 0.1	0.7 ± 0.04	1.9 ± 0.1	1.7 ± 0.1	0.8 ± 0.1	ND†	0.2 ± 0.03	0.9 ± 0.07	ND	ND
Pb	ND	ND	ND	ND	0.8 ± 0.1	0.5 ± 0.1	ND	ND	ND	ND
Ni	9.6 ± 0.2	3.3 ± 0.1	8.5 ± 0.1	4.3 ± 0.1	5.2 ± 0.1	5.3 ± 0.1	4.3 ± 0.2	10.9 ± 0.1	7.4 ± 0.1	1.4 ± 0.1
Cr	4.6 ± 0.1	2.5 ± 0.1	1.8 ± 0.1	2.1 ± 0.1	5.8 ± 0.1	5.3 ± 0.1	0.1 ± 0.04	ND	ND	ND
Fe	688.2 ± 0.4	491.1 ± 0.4	30.0 ± 0.5	399.9 ± 0.5	426.0 ± 36	419.0 ± 3.8	316.7 ± 1.9	46.8 ± 0.2	386.1 ± 0.6	49.9 ± 0.1
Mn	52.5 ± 0.4	20.6 ± 0.3	7.9 ± 0.1	17.9 ± 0.1	28.8 ± 0.2	23.8 ± 0.8	42.3 ± 0.3	37.7 ± 0.1	37.2 ± 0.7	30.6 ± 0.2
Zn	24.0 ± 0.1	19.8 ± 0.1	17.5 ± 0.1	10.5 ± 0.1	57.7 ± 0.3	56.6 ± 1.3	25.4 ± 0.3	17.6 ± 0.2	51.9 ± 0.5	29.6 ± 1.9

*Expressed in $\mu g\ g^{-1}$, mean ± SD.
ND, not determined.

data. In the precision test, the average RSD % for all investigated trace metals are in the range of 1–10% ($n = 10$). The range of the calibration standards for copper, iron, nickel, cadmium, manganese, lead, chromium and zinc on flame atomic absorption spectrometric determinations were 0.0–5.0, 0.0–5.0, 0.0–5.0, 0.0–2.0, 0.0–5.0, 0.0–10.0, 0.0–10.0 and 0.0–1.0 mg L^{-1} respectively. The correlation coefficient of the calibration curves were generally 0.999.

The concentration of traces heavy metals in investigated plant samples from western Anatolia, Turkey are given in Tables 2 and 3 respectively. The values are given as mean ± SD. The results are means of three replicates. The metal levels determined were based on plants' dry weight. While cadmium is the lowest level in the all herbal samples from western Anatolia with some exceptions, the levels of iron in the samples were generally the highest.

Copper is one of the essential micronutrients, and its adequate supply for growing plants should be ensured through artificial or organic fertilnrs (Itanna, 2002). Copper ranged from 3.8 $\mu g\ g^{-1}$ in leaf of laurel (*Laurus nobilis*) to 35.4 $\mu g\ g^{-1}$ in leaf of mullein (*Verbascum* spp.). Copper levels of basil (*Ocimum basilicum* L.), rosemary (*Rosmarinus officinalis* L.), laurel (*Laurus nobilis* L.) and lavender (*Lavandula officinalis* L.) from Konya, Turkey has been reported as 8.05, 6.66, 3.17 and 10.7 $\mu g\ g^{-1}$ respectively by Ozcan (2004). In our work the copper levels of basil, rosemary, laurel and lavender were found as 20.1, 9.2, 3.8 and 11.3 $\mu g\ g^{-1}$ respectively.

Cadmium is a nonessential element in foods and natural waters, and it accumulates principally in the kidneys and liver. Cadmium in foods is mostly derived from various sources of environmental contamination (Adriano, 1984). Concentrations of cadmium were generally low in nearly all analysed samples, ranging from 0.2 to 2.7 $\mu g\ g^{-1}$; lowest in yarrow herb (*Achillea millefolium*) and highest in leaf of laurel (*Laurus nobilis*). Cadmium content of yarrow herb (*Achillea millefolium*) from Austria has been reported as 0.49 $\mu g\ g^{-1}$ by Chizzola *et al*. (2003).

Lead is widely distributed in spices and herbal plants. Lead, being a serious cumulative body poison, enters into the body system through air, water and food, and cannot be removed by washing the fruits and vegetables (Divrikli *et al*., 2003, De Leonardis *et al*., 2000). The lowest concentration of Pb was in the leaf of laurel (*Laurus nobilis*) (0.1 $\mu g\ g^{-1}$), while the highest level was in root of chard (2.8 $\mu g\ g^{-1}$). Lead in rosemary (*Rosmarinus officinalis* L.) from Austria was given as 1.45 $\mu g\ g^{-1}$ (Chizzola *et al*., 2003). The concentration of the cadmium and lead in leaf of chard (*Beta vulgaris* L. var. cicla) were given by Nadal *et al*. (2004) as Cd 0.09 $\mu g\ g^{-1}$, Pb 0.71 $\mu g\ g^{-1}$. Lead level of basil, rosemary and lavender from middle Anatolia, Turkey has been reported as 2.10, 8.36 and 1.13 $\mu g\ g^{-1}$ (Ozcan, 2004).

Nickel also plays some roles in body functions including enzyme functions. Nickel occurs naturally more in plants than in animal flesh. In very trace amounts, it may be beneficial to activate some enzyme systems, but its toxicity at higher levels is more prominent. However, nickel toxicity in humans is not a very common occurrence because the absorption of nickel is very low (Onianwa *et al*., 2000). The nickel contents in the samples were in the range of 1.4–11.3 $\mu g\ g^{-1}$ (Tables 2 and 3). The lowest level of nickel was in root of centaury (*Centaurium erythraea*) and the highest was in leaf of laurel (*Laurus nobilis*). The level of nickel in the leaf of rosemary, laurel leaf, chard, marjoram and sheep sorrel samples were higher than those in the roots. This point can be explained by the transportation of nickel from the roots to the leaf of the samples. The nickel range of some herbal teas from middle Anatolia was reported as 11.3–37.0 $\mu g\ g^{-1}$ by Colak *et al*. (2005). Cala *et al*. (2005) reported concentration range of nickel of *Rosmarinus officinalis* L. samples from Madrid, Spain as 0.65–1.85 $\mu g\ g^{-1}$. Our

nickel values are in agreement with those reported in the literature (Cala *et al*., 2005; Colak *et al*., 2005).

Chromium levels in all the samples were in the range of 0.1–9.7 $\mu g\ g^{-1}$ (lowest in yarrow, highest in marjoram). The concentrations of chromium in basil, rosemary, laurel and lavender from Konya, Turkey was reported as 7.95, 8.93, 11.0 and 19.0 $\mu g\ g^{-1}$ respectively by Ozcan (2004). The concentration of chromium in leaf of chard (*Beta vulgaris* L. var. cicla) was reported as 0.48 $\mu g\ g^{-1}$ by Nadal *et al*. (2004). Mean chromium content of basil samples (*Ocimum basilicum*) from Spain was reported as 0.54 $\mu g\ g^{-1}$ (0.54–0.51 $\mu g\ g^{-1}$) (Garcia *et al*., 2000). In laurel samples from Spain (Garcia *et al*., 2000), the concentration of chromium was reported in the range of 0.41–0.68 $\mu g\ g^{-1}$ (mean: 0.60 $\mu g\ g^{-1}$). In our work, chromium content of leaf and root of laurel (*Laurus nobilis*) were 2.7 and 4.0 $\mu g\ g^{-1}$ respectively (Table 2).

Of all the micronutrients, iron is required by plants in the largest amount. The iron contents in the samples were in the range of 30.0 $\mu g\ g^{-1}$ in leaf of mullein 945.3 $\mu g\ g^{-1}$ in leaf of basil (Tables 2 and 3). Generally, the analysed samples contained, relative to the other trace metals, higher concentrations of iron. The concentrations of iron in rosemary (*Rosmarinus officinalis* L.) samples from Konya, Turkey (Chizzola *et al*., 2003; Ozcan, 2004) were given as 547 and 375.2 $\mu g\ g^{-1}$ respectively. Iron content of basil sample from Konya,Turkey was 225.68 $\mu g\ g^{-1}$ (Ozcan *et al*., 2005), which can be compared for iron value for basil from western Anatolia.

Manganese activates numerous essential enzymes. Food contains trace amounts of manganese (Colak *et al*., 2005). The highest concentrations of manganese were found in the leaf of laurel (152.5 $\mu g\ g^{-1}$). The lowest level of Mn was in mullein flower (7.9 $\mu g\ g^{-1}$). Mean manganese level in leaf of chard (*Beta vulgaris* L. var. cicla) was reported as 81.7 $\mu g\ g^{-1}$ by Nadal *et al*. (2004). Manganese level of *Ocimum basilicum* L., *Rosmarinus officinalis* L., *Laurus nobilis* L. and *Lavandula officinalis* L. have been reported as 117, 41.2, 32.6 and 50.1 $\mu g\ g^{-1}$ repectively. Our manganese values are in agreement with those reported in the literature for herbal samples (Chizzola *et al*., 2003; Colak *et al*., 2005).

Zinc is one of the important metals for normal growth and development in human beings. Deficiency of zinc can result from inadequate dietary intake, impaired absorption, excessive excretion or inherited defects in zinc metabolism (Colak *et al*., 2005; Narin *et al*., 2005). Zinc deficiency is of growing concern in the developing world because of the consumption of plant foods that have inhibitory components for zinc absorption. Especially, in these populations, zinc deficiency is related to the high consumption of bread made without yeast. Zinc in our samples was detected in the range of 5.2–83.7 $\mu g\ g^{-1}$. While the highest level of zinc was in leaf of marjoram, the lowest level was in root of laurel. Mean zinc content of basil (*Ocimum basilicum L.*), rosemary (*Rosmarinus officinalis* L.), laurel (*Laurus nobilis* L.) and lavender (*Lavandula officinalis* L.) from Konya, Turkey was reported as 13.7, 15.6, 21.9 and 12.3 $\mu g\ g^{-1}$ respectively by Ozcan (2004).

The recommended dietary allowances (RDA) as mg day^{-1} $person^{-1}$ for copper, iron, zinc and manganese are 2, 18, 15 and 5 respectively (National Research Council, 1989). The levels of copper, iron, zinc and manganese in the samples were lower than the Food and Nutrition Board of the National Academy of Sciences, United States recommendations given above.

Although there are no RDA value for cadmium, lead, nickel and chromium, some values were given by Food and Nutrition Board of the National Academy of Sciences, United States and other authorities. The recommended daily intake for chromium is 0.20 mg day^{-1} (National Research Council, 1989). Provisional tolerable intake for lead and cadmium is 0.21 and 0.06 mg day^{-1} respectively (WHO, 1993). Average daily intake from food for nickel is 0.30 mg day^{-1} (WHO, 1996). The levels of Cd, Pb, Ni and Cr were also lower than the levels given above by World Health Organization and Food and Nutrition Board of the National Academy of Sciences-United States.

Conclusion

The levels of the investigated heavy metal ions studied compared well with levels in herbal plants and spice samples from other parts of the world. Generally, lower levels of the investigated metals were found in the roots of the samples than in the leaf of the samples, because of the transportation of the metals from the roots to leaf. In order to evaluate the convenience of including foods in diets, metal levels of food samples can be useful as nutritional guidances. Also the values in the present work for the levels of the traces metal ions in the herbal plants and spice samples from western Anatolia, Turkey could help in the food composition tables for Turkish people.

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