Research

Determination of metal contents in some green leafy vegetables in Marmara region of Turkey

Medet Zor[1] · **Sevgi Kocaoba**[1]

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Abstract

This study focused on the analysis of mineral and heavy metal contents of spinach, lettuce and parsley samples in different regions to investigate the risks of chronic exposure to heavy metal contamination. After the microwave digestion process, the metal contents of the samples (Na, Mg, Al, P, K, Ca, Fe, Cu, Zn, As, Se, Cd, Sn, Hg, Pb) were determined with the ICP-MS. Two parallel analysis samples were prepared from each sample, analyzed three times, and the mean values of the analyzes performed were reported. From the analysis, the order of finding heavy metals in the samples is Cu > Pb > Cd > As > Sn > Hg. The results were evaluated statistically and Cu, Pb, Cd, As and Sn values were found high in spinach samples ($p < 0.05$). According to the results of the analysis; the samples with the highest content of Cu, Pb and Cd in spinach samples are samples taken from Tekirdağ (5.1 ± 0.3 mg/kg), Edirne (0.106 ± 0.007 mg/kg) and Tekirdağ (0.080 ± 0.004 mg/kg), respectively. One of the important reasons for this is the increase in industrial areas in those regions. The results were compared with the certified reference materials, the quantities of analytes were determined to be compatible with the certificate values and the results of the analysis were proved to be correct.

Article Highlight

- Transfer of trace elements to plants poses a risk of pollution for the ecosystem and livestock.
- This study focused on the analysis of mineral and heavy metal contents of spinach, lettuce and parsley samples in different regions.
- Heavy metal levels were determined with ICP-MS and the results were compared with the certified reference materials.
- Agricultural practices with amendments can be used to reduce heavy metal levels in vegetables.

Keywords Green leafy vegetables · Heavy metals · Microwave digestion · ICP-MS · Standart reference material · Food pollution

1 Introduction

Environmental pollution has increased significantly with the increase of urbanization and industrialization. Solid and liquid wastes are the main factors that cause soil pollution. These wastes accumulate in the soil and pass into groundwater, causing them to become contaminated. Even if heavy metals are trace levels, solid and liquid wastes have an important place due to their toxicity and accumulation properties [1–7]. Soil pollution

✉ Sevgi Kocaoba, kocaobasevgi@hotmail.com | [1]Department of Chemistry, Faculty of Art and Science, Yildiz Technical University, Davutpasa Cad. No: 127, 34220 Istanbul, Turkey.

is widely dispersed due to environmental impacts and urbanization and industrialization around it. Elements such as Cr, Pb, Cd, As, Hg, which have an important place in heavy metal pollution, are extremely effective in soil and water pollution. These heavy metals spread to the environment as a result of their mining activities, production of electronic equipment and their subsequent wastes, fertilizers and pesticides used in agricultural areas, sewage wastes, paint and other industries [8–13]. Then they enter the feeding system of humans and animals in various ways. Heavy metals are taken into the organism by mouth, respiration and skin and most of them cannot be excreted by the body's excretory pathways (kidney, liver, intestine, lung, skin) without special support. Therefore, most of the heavy metals accumulate in biological organisms. Whether a heavy metal is vital depends on the organism considered. For example, nickel has toxic effects on plants, while it is an element that must be present in traces in animals. There are some basic and essential elements like iron, cobalt, zinc, copper, magnesium for both humans and animals. But when taken in excess, they are harmful to human health. For example, copper is indispensable for red blood cells. It is an essential element for many oxidation and reduction reactions in humans and animals, but the excess of copper causes irritation in the nose, mouth, and eyes. In addition, dizziness, headache, stomach pain and nausea and diarrhea occur [14, 15]. Zinc is a very necessary metal in terms of nutrition [16, 17]. As a result of its insufficiency, significant health problems occur. On the other hand, if exposure to excessive amounts of zinc is rarely known, gastrointestinal system disorders and diarrhea occur. When exposed to cobalt excessively, it causes toxic symptoms such as vasodilation, flushing and cardiomyopathy. Mercury, lead, chromium, cadmium, and arsenic have been the most common heavy metals that induced human poisonings. Here, we reviewed the mechanistic action of these heavy metals according to the available animal and human studies. Acute or chronic poisonings may occur following exposure through water, air, and food. The ORD (mg/kg person/day)(An estimated exposure of metal to the human body per day associated with no potential hazardous effect during lifetime) for Pb, Co, Cd, Zn, Cu, and Mn used were 0.004, 0.02, 0.001, 0.3, 0.02, 0.04, and 0.033 mg/kg person/day, respectively [18–24].

Green leafy vegetables are the most nutritious in foods. It is very rich in minerals (iron, calcium, potassium and magnesium), vitamins (K, C, E and many types of B vitamins) and phyto-nutrients (beta-carotene, lutein and zeaxanthin) [25]. They protect our cells from aging and being damaged. They also contain a significant amount of fibers. However, the content of the soil in which these plants are grown is extremely important. The heavy metal contents of the plants grown in the lands where the environmental pollution is intensive can also exceed the permissible levels. In addition, residues of fertilizers and pesticides used are found on the plants grown. Where vegetables are grown, the metals in the soil can be risked for human health by taking them through food, inhaling the soil dust and accidentally ingesting it by hand-to-mouth behavior [26–33].

Food and drinks taken by humans are an important way of exposure to important trace elements in terms of toxicity and nutrition. In addition, they have the property of accumulating in parts of the body such as brain, liver and bones after being taken through food and drinks. Due to these effects, it is important to monitor and identify these trace metal types in food samples. There are several methods to determination of trace metal ions, atomic spectrometry is often used. These methods are flame atomic absorption spectrometry (FAAS), electrothermal atomic absorption spectrometry (ETAAS), atomic fluorescence spectrometry (AFS), inductively coupled plasma optical emission spectroscopy (ICP-OES), etc. [34–44].

This study focused on the mineral and heavy metal levels in these vegetables to investigate possible risks in the population in different regions when they are exposed to chronic heavy metal contamination of spinach, lettuce and parsley, the most consumed vegetables. Vegetable samples were taken for 54 samples to be examined from the Marmara Region and sampling areas were represented by the population.

2 Materials and methods

2.1 Materials

In this study, the population of the provinces in the Marmara Region was taken into consideration in the selection of the samples taken for analysis. Thus, it was thought that a more accurate table would emerge by selecting samples based on the consumption of vegetables. The population of the places where the samples are taken is generally around 1 million. The places where the samples were taken and their populations are given in Table 1. The sample collection locations are shown on the map and given in Fig. 1.

Since the number of samples to be analyzed is limited to 18, samples have not been taken from some provinces and districts and combined with neighboring provinces and districts.

Table 1 The places where the samples were taken and their populations

No	Place	Content	Population
1	Balıkesir–Center	Balıkesir and Çanakkale	1,640,750
2	Bilecik–Center	Bilecik	203,849
3	Bursa–Center	Osmangazi and Yıldırım	1,442,417
4	Bursa–Nilüfer	Yalova and other districts of Bursa	1,416,244
5	Edirne–Center	Edirne, Kırklareli	739,515
6	İstanbul–Esenler	Bağcılar, Güngören	1,510,167
7	İstanbul–Esenyurt	Büyük Çekmece, Beylikdüzü, Başakşehir, Silivri	1,055,771
8	İstanbul–Eyüp	Çatalca, Sultangazi, Gaziosmanpaşa	1,374,947
9	İstanbul–Fatih	Beyoğlu, Zeytinburnu	970,785
10	İstanbul–Kadıköy	Ataşehir, Üsküdar, Maltepe, Adalar	1,917,663
11	İstanbul–Küçükçekmece	Bakırköy, Avcılar	1,315,511
12	İstanbul–Pendik	Kartal, Tuzla, Şile	1,276,499
13	İstanbul–Şişli	Kâğıthane, Beşiktaş	933,471
14	İstanbul–Sarıyer	Bahçelievler, Arnavutköy	1,086,439
15	İstanbul–Ümraniye	Çekmeköy, Beykoz, Sancaktepe, Sultanbeyli	1,627,580
16	Kocaeli–Center	Kocaeli	1,601,720
17	Sakarya–Center	Sakarya	888,556
18	Tekirdağ–Center	Tekirdağ	829,873

2.2 Methods

In this study, it was aimed to measure the mineral and heavy metal levels of spinach, lettuce and parsley, which are the most consumed vegetables from different regions, in 54 different samples. For this purpose, NMKL (Nordic Committee on Food Analysis. No: 186, 2007) method used to detect metal residues in foodstuffs was applied. The samples were dissolved in the microwave oven (microwave oven-Berghof MWS–3 +) by wet combustion method. The metal contents of samples were measured by ICP-MS (Agilent 7500 cx) device according to certain standards. The operating parameters of the ICP-MS used in the determination of metals are given in Table 2.

2.3 Chemicals

High purity single element standards of Na, K, Ca, Mg, Al, Se, Cu, Zn, Fe, Pb, Cd, Sn, As, Hg were supplied from Merck. Ultrapure quality HNO_3 (65%), HCl (30%), H_2O_2 (31%) (Merck) were used for the experiments. The solutions used

Fig. 1 The sample collection locations in Marmara Region of Turkey

Table 2 Operating conditions of the used Agilent 7500cx Brand ICP–MS device

		Recommended value	Recommended range
Plasma parameters	RF–Power (W)	1500	1400–1650
	Sample depth (mm)	8	7–10
	Carrier Gas Flow (l/min)	0.91	0.8–1.0
	Make up Gas Flow (l/min)	0.17	0.1–0.3
	Nebulizer Pump (rps)	0.1	0.05–0.25
	Spray Chamber Temperature (°C)	2	2.0–2.2
Detector parameters	Discriminator (mV)	8.0	–
	Analog HV	1770	–
	Pulse HV	1010	–

in all studies were prepared fresh from stock solutions with Milli-Q water.

2.4 Preparation of samples

Vegetable samples were washed and cleaned with distilled water and left for 2 h to allow water to flow on them. Then, the samples were homogenized in the shredder and taken into the sample containers. The samples were weighed with a sensitivity of 1 mg, in two parallels to the microwave weighing containers, between 0.2 and 0.5 g. By transferring microwave weighing cups to the incinerator, 4 ml ultrapure HNO_3 (65%) and 1 ml H_2O_2 (30%) combustion chemicals were added. One blind sample containing the same quantities of combustion chemicals was prepared. After waiting for 15 min, the combustion vessels were closed and placed in the microwave device and were broken down at high temperature and pressure. When the combustion is complete, after the containers have cooled, their lids are opened carefully and the contents of the vessel are filtered into a 50 ml flask. The device (ICP-MS) was added with a solution of 675 µl of HCl, 0.5% HCl in accordance with the operating conditions. The obtained filtrate was completed to 50 ml. Subsequently, blinds and samples were filtered through Millipore Millex—HV (Hydrophilic PVDF 0.45 µm) membrane filter into vials to be given to the device.

2.5 Preparation of CRM sample

In order to verify the analyzes, the sample "Dried Tomato" was used as "Certified Reference Material (CRM)". The analysis steps of this sample are as follows;

The samples were weighed between 0.2 and 0.5 g in microwave weighing pans with two parallels with 1 mg sensitivity. Microwave weighing vessels were transferred to the combustion vessel, and 4 ml ultrapure HNO_3 (65%) and 1 ml H_2O_2 (30%) combustion chemicals were added, and a blind sample containing the same quantities of combustion chemicals was prepared. It was placed in the microwave device by turning it off and it was ensured that it was broken down at high temperature and pressure in a suitable burning program.

2.6 Determination of samples

After dissolved in the microwave combustion system, the solutions were analyzed with ICP-MS, and the amounts of Al, As, Cu, Hg, Zn, Fe, Pb, Cd, Sn, Ca, Mg, P, K, Se and Na were determined in mg/kg.

3 Results and discussion

As a result of 324 analyzes on 54 samples, metal contents were calculated for spinach, lettuce and parsley. In addition to the analyzes, verification was made by working with certified reference materials (CRM).

The metal contents in the sample were calculated from the calibration curve drawn according to the "ratio" value corresponding to the concentration of the analytical standard. The amount in the sample was found using the formula below;

$$Xs = (Cs - Cb) \times (V/m)/1000 \quad (1)$$

$$\text{The amount of the element in the sample (mg/kg)} = (\text{sample value} - \text{blind value}) \times (\text{dilution volume/sample weight})/1000$$

The amount of various elements in spinach, lettuce and parsley samples were determined by calculating the slope formula of the calibration curve. Sources of uncertainty regarding the relevant results were also identified. Sources of uncertainty; stock solutions, dilution coefficient, calibration curve, repeatability and uncertainties from recovery. Uncertainties about these have been calculated.

Measurement results are reported as follows:

$$X \pm U(mg/L)(95\%k = 2) \quad (2)$$

X = The value calculated from the calibration curve after the measurement; U = Extended uncertainty.

Table 3 LOQ and calibration interval values of the analyzed elements

Element	LOQ (µg/l)	Calibration range (µg/l)	Linearity (R^2)
Aluminium (Al)	70	0–375	> 0.999
Arsenic (As)	1	0–10	> 0.999
Cupper (Cu)	70	0–375	> 0.999
Mercury (Hg)	1	0–10	> 0.999
Zinc (Zn)	50	0–375	> 0.999
Iron (Fe)	70	0–375	> 0.999
Phosphorus (P)	120	0–50,000	> 0.999
Cadmium (Cd)	2	0–10	> 0.999
Tin (Sn)	60	0–375	> 0.999
Calcium (Ca)	170	0–50,000	> 0.999
Lead (Pb)	1	0–10	> 0.999
Magnesium (Mg)	320	0–50,000	> 0.999
Potassium (K)	260	0–50,000	> 0.999
Selenium (Se)	1	0–10	> 0.999
Sodium (Na)	150	0–50,000	> 0.999

LOQ and calibration interval values of the analyzed elements are given in Table 3. Metal concentrations in spinach, lettuce and parsley samples are given in Figures between 2 and 10 (Figs. 2, 3, 4, 5, 6, 7, 8, 9 and 10).

Vegetable samples were collected from the Marmara Region and the places to be sampled were determined according to the city population. If we look at the metal levels in the samples we examined;

Metal levels in spinach samples; Na concentrations between 57 and 1283 mg/kg; Mg concentrations between 528 and 1270 mg/kg; P concentrations between 197 and 513 mg/kg; K concentrations between 3408 and 7393 mg/kg; Ca concentrations between 636 and 2330 mg/kg; Al concentrations between 12.5 and 100.2 mg/kg; Fe concentrations between 12.4 and 120.1 mg/kg; Cu concentrations between 0.61 and 28.3 mg/kg; Zn concentrations between 3 and 10.1 mg/kg; Sn concentrations between 0.06 and 0.145 mg/kg; As concentrations between 0.017 and 0.05 mg/kg; Se concentrations between 0.001 and 0.045 mg/kg; Cd concentrations between 0.025 and 0.080 mg/kg; Hg concentrations < 0.001; Pb concentrations between 0.005 and 0.095 mg/kg.

Metal levels in lettuce samples; Na concentrations between 48 and 663 mg/kg; Mg concentrations between 116 and 745 mg/kg; P concentrations between 119 and 345 mg/kg; K concentrations between 1860 and 4135 mg/

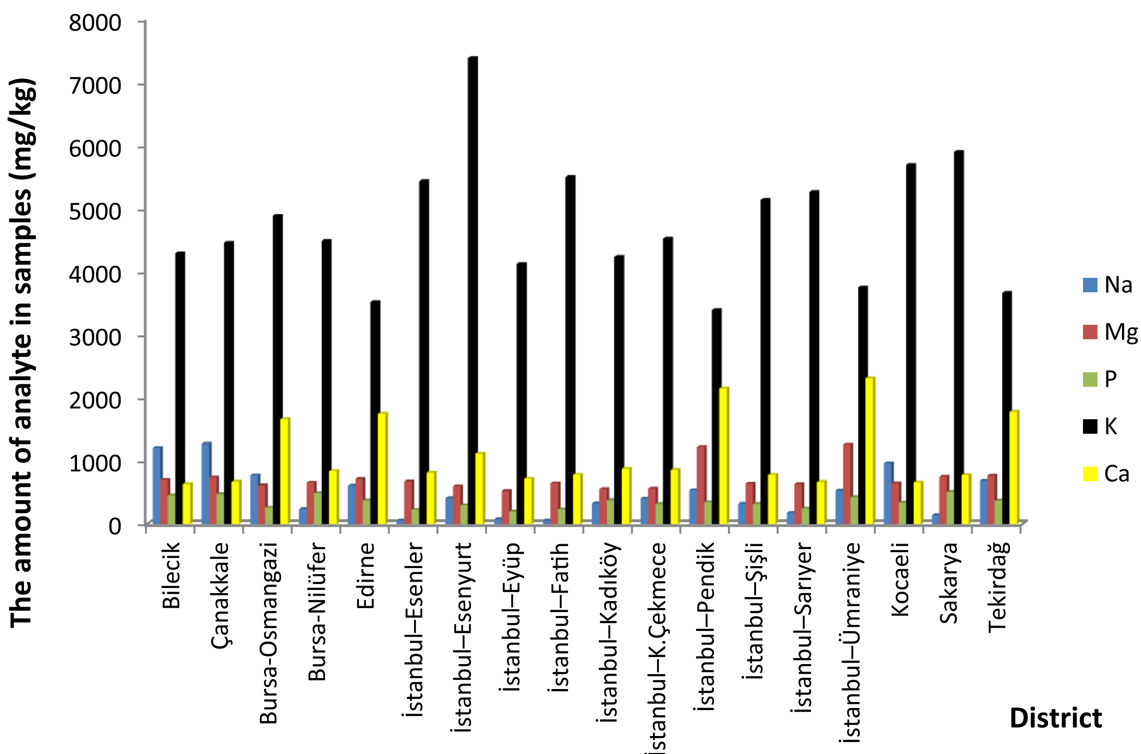

Fig. 2 Na, Mg, P, K and Ca analysis results in spinach samples (mg/kg)

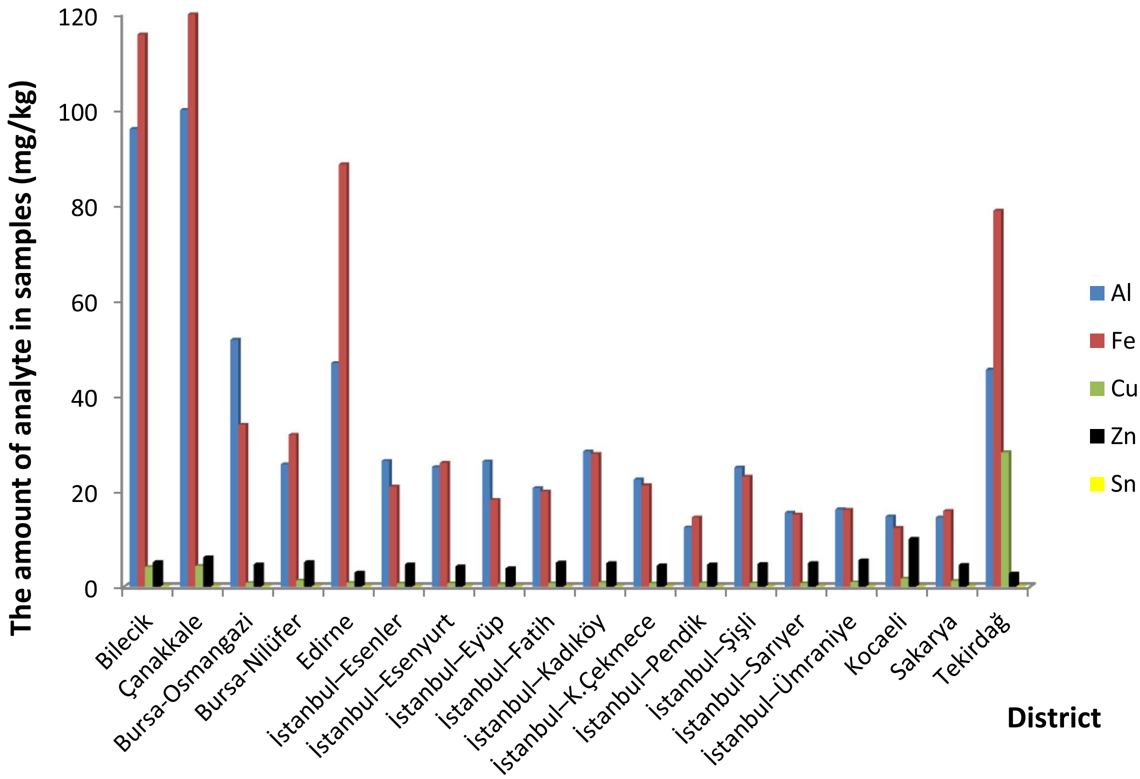

Fig. 3 Al, Fe, Cu, Zn and Sn analysis results in spinach samples (mg/kg)

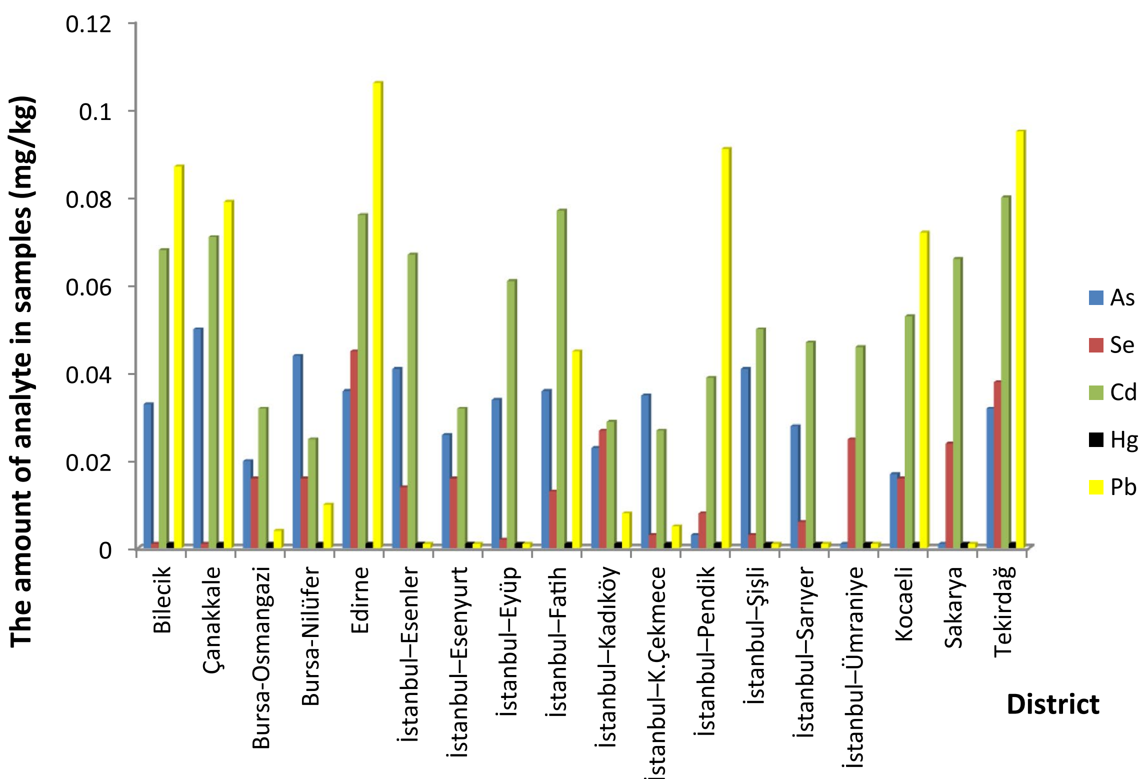

Fig. 4 As, Se, Cd, Hg and Pb analysis results in spinach samples (mg/kg)

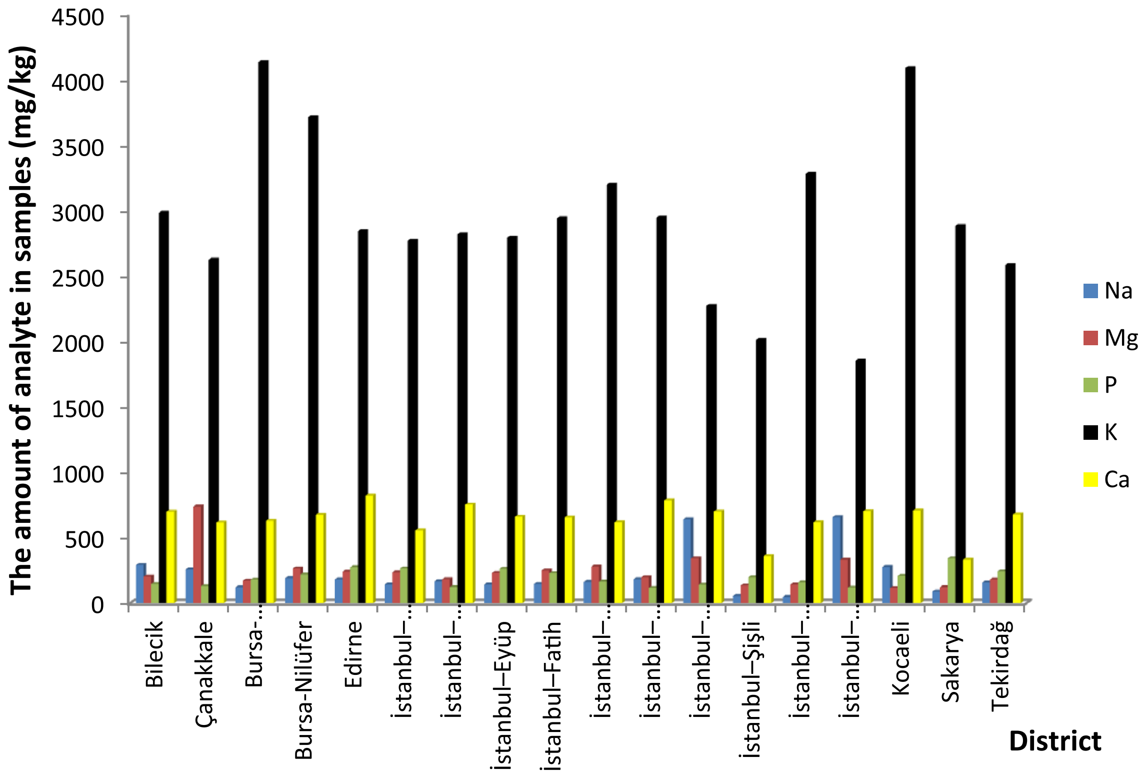

Fig. 5 Na, Mg, P, K and Ca analysis results in lettuce samples (mg/kg)

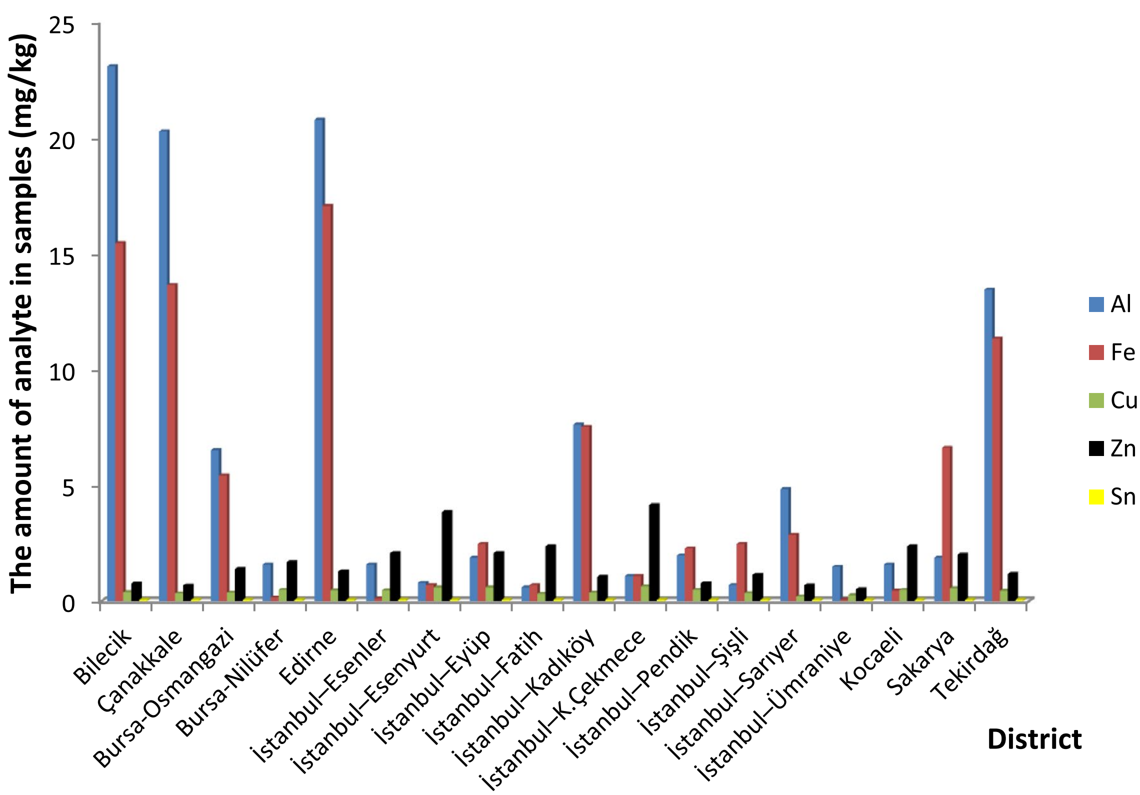

Fig. 6 Al, Fe, Cu, Zn and Sn analysis results in lettuce samples (mg/kg)

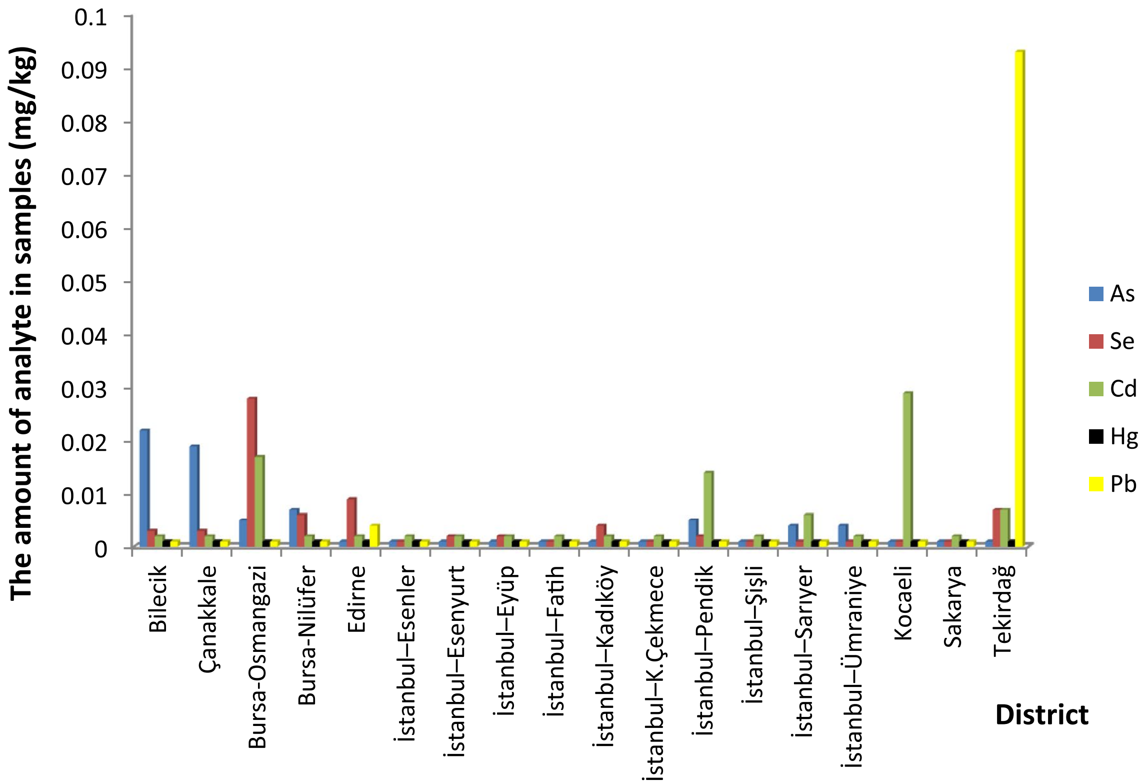

Fig. 7 As, Se, Cd, Hg and Pb analysis results in lettuce samples (mg/kg)

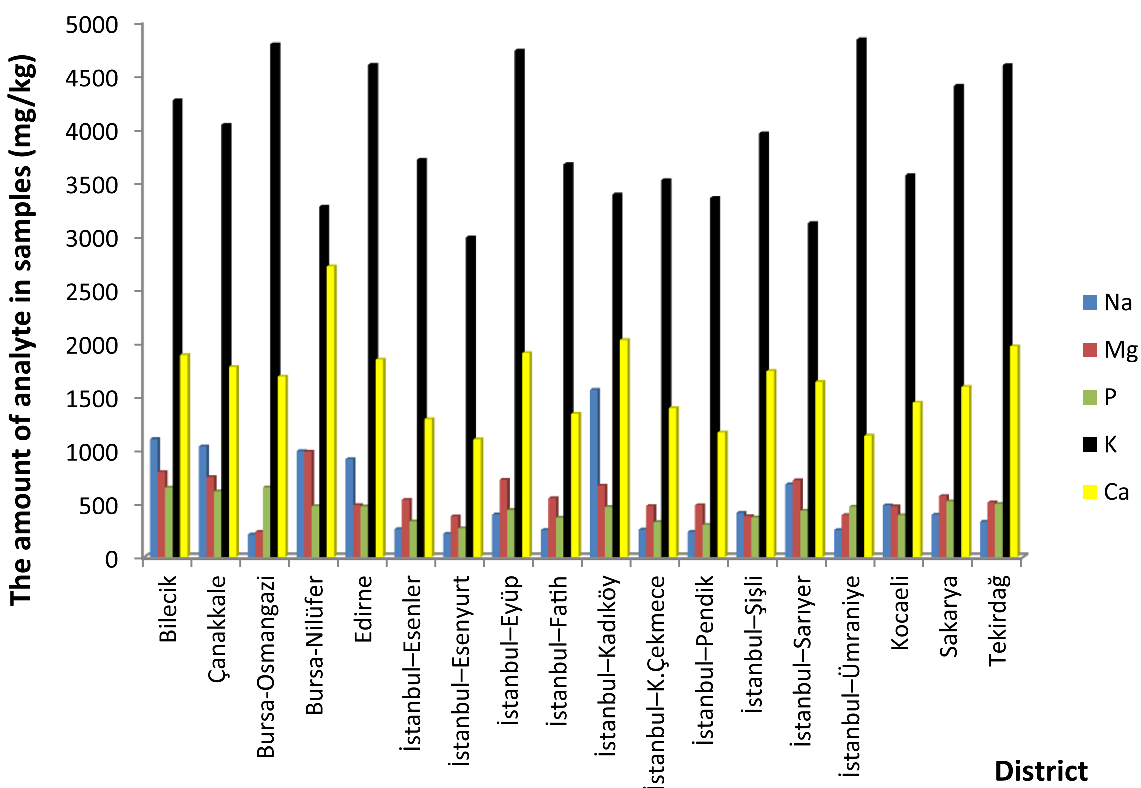

Fig. 8 Na, Mg, P, K and Ca analysis results in parsley samples (mg/kg)

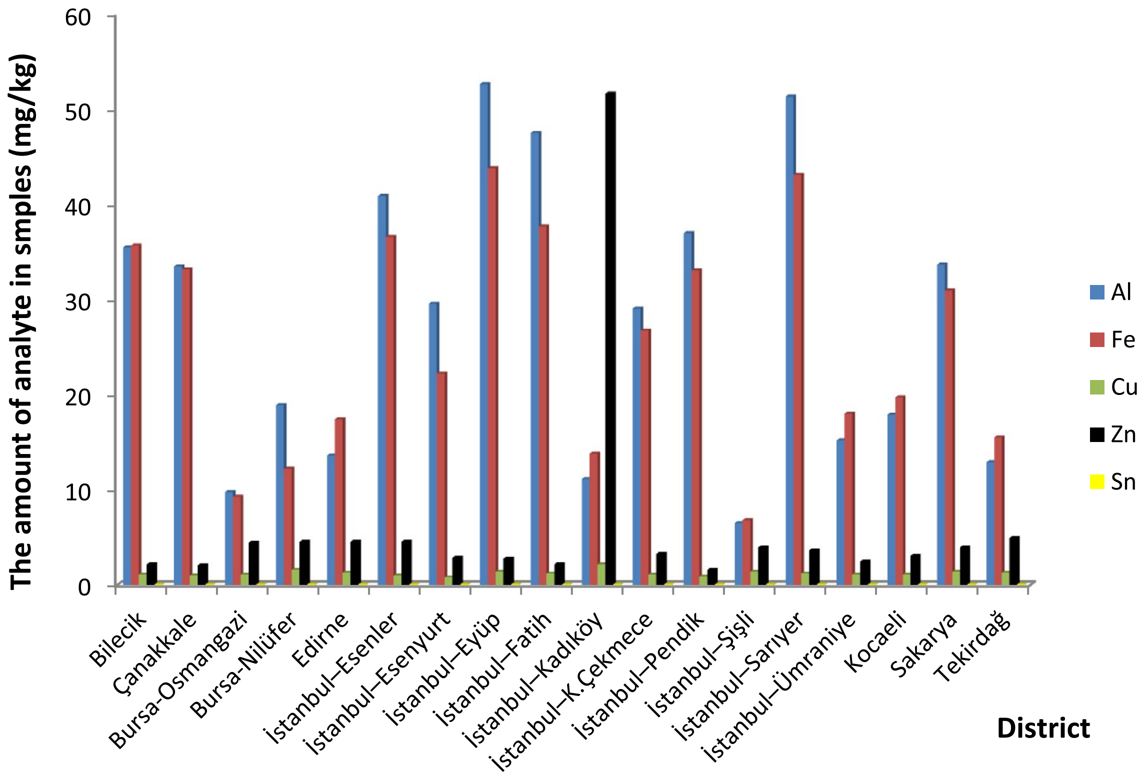

Fig. 9 Al, Fe, Cu, Zn and Sn analysis results in parsley samples (mg/kg)

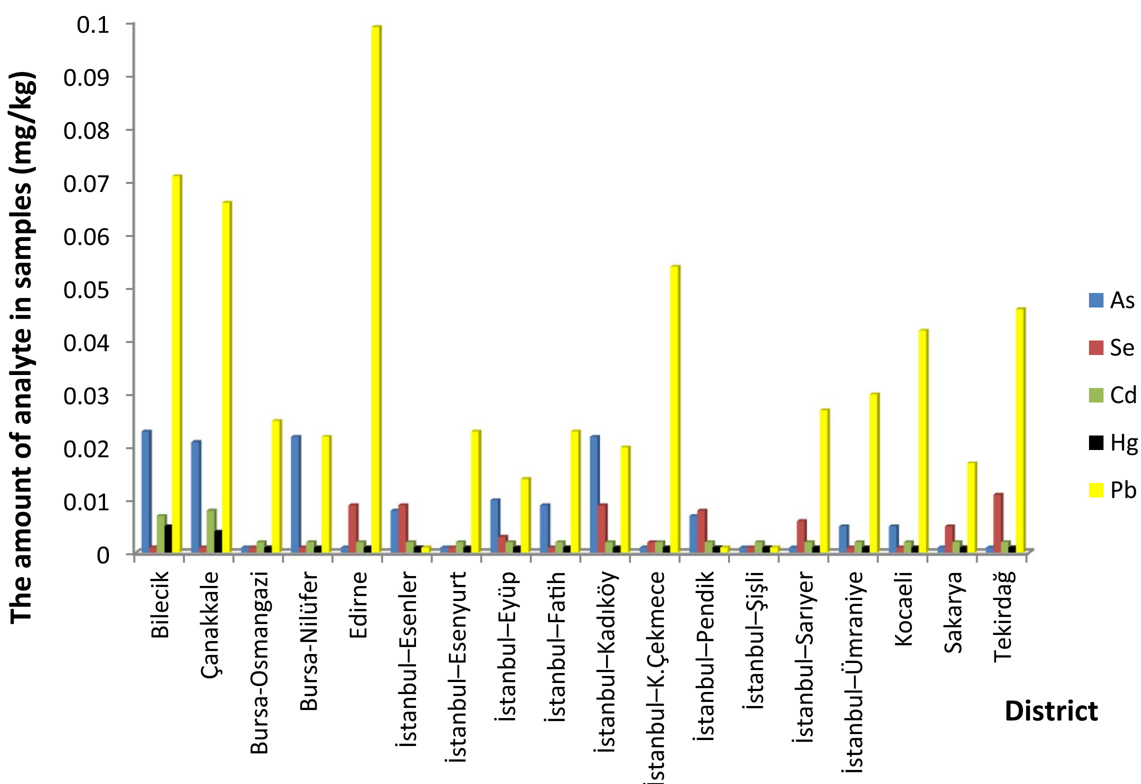

Fig. 10 As, Se, Cd, Hg and Pb analysis results in parsley samples (mg/kg)

kg; Ca concentrations between 335 and 760 mg/kg; Al concentrations between 0.6 and 23.1 mg/kg; Fe concentrations between 0.16 and 17.1 mg/kg; Cu concentrations between 0.20 and 0.64 mg/kg; Zn concentrations between 0.52 and 4.20 mg/kg; Sn concentrations < 0.06 mg/kg; As concentrations between 0.001 and 0.022 mg/kg; Se concentrations between 0.002 and 0.028 mg/kg; Cd concentrations between 0.002 and 0.029 mg/kg; Hg concentrations < 0.001; Pb concentrations between 0.001 and 0.093 mg/kg.

Metal levels in parsley samples; Na concentrations between 210 and 1565 mg/kg; Mg concentrations between 236 and 989 mg/kg; P concentrations between 302 and 651 mg/kg; K concentrations between 2988 and 4837 mg/kg; Ca concentrations between 1103 and 2719 mg/kg; Al concentrations between 6.6 and 47.6 mg/kg; Fe concentrations between 6.9 and 43.9 mg/kg; Cu concentrations between 0.8 and 1.6 mg/kg; Zn concentrations between 1.6 and 51.7 mg/kg; Sn concentrations < 0.06 mg/kg; As concentrations between 0.001 and 0.023 mg/kg; Se concentrations between 0.001 and 0.011 mg/kg; Cd concentrations between 0.002 and 0.008 mg/kg; Hg concentrations between 0.001 and 0.005; Pb concentrations between 0.001 and 0.071 mg/kg.

According to the literature information, when it is evaluated in terms of the average of the analyzes;

It can be said that As, Cu, Cd and Pb elements are close to some values in the literature in spinach, lettuce and parsley samples [13, 16, 33, 45–51]. Comparative data from literature for the determination of some heavy metals in some food samples were given at Table 4.

It is thought that the differences observed in the heavy metal content of vegetables are due to the characteristic features of the vegetables. However, huge differences can be observed between the places where the samples are taken. It is thought that this difference may arise from the differences in the environmental conditions in which vegetables are grown and similar factors. In areas where the industry is more concentrated, heavy metal levels are slightly higher.

When examined in terms of heavy metals, it was observed that the most accumulated heavy metal was lead and this element was found in the most spinach. Similar results have been obtained in previous studies in different countries [52–57]. When evaluated in terms of

Table 4 Comparative data from literature for the determination of some heavy metals in some food samples

Vegetables	Metal (mg/kg)							References
	Cu	Zn	Fe	Pb	Cd	As	Hg	
Spinach	0.61–28.3	3–10.1	12.4–120.1	0.005–0.095	0.025–0.080	0.017–0.05	< 0.001	This study
Lettuce	0.20–064	0.52–4.20	0.16–17.1	0.001–0.093	0.002–0.029	0.001–0.022	< 0.001	
Parsley	0.8–1.6	1.6–51.7	6.9–43.9	0.001–0.071	0.002–0.008	0.001–0.023	0.001–0.005	
Spinach			6					[16]
Lettuce			1.1					
Parsley								
Spinach	0.97	20.81		0.97	0.51	0.31		[33]
Lettuce	0.77	11.79		1.16	0.46	0.66		
Parsley								
Spinach								[46]
Lettuce	45.4	218.3		4.4	0.34			
Parsley	35.43	259.2		6.5	0.4			
Spinach	2.45	2.99	21.5	0.13	0.043	0.014		[52]
Lettuce	0.17	1.01	4.04	0.013	0.038	0.007		
Parsley	0.29	0.88	7.20	0.025	0.002	0.004		
Spinach	6.29	246.7		0.62	0.36			[53]
Lettuce	5.67	51.24		0.06	0.14			
Parsley								
Spinach	0.99–1.7	3.99–14.00	13.02–127.6	0.207–1.008	0.055–nd			[58]
Lettuce	0.32–0.71	1.86–3.63	4.04–66.13	0.075–0.249	0.031–nd			
Parsley	1.19–2.83	2.12–16.82	22.59–181.7	0.005–0.077	nd			
Spinach								[59]
Lettuce	7.95–8.38	28.78–38.53	239.0–684.5	0.53–0.94	0.24–0.30			
Parsley	6.87–7.54	21.93–26.06	861.5–1129.0	0.84–2.27	0.49–0.70			

vegetable types as a result of the statistical analysis, it can be said that the differences in many elements are found to be important at the level of 0.05, and the results obtained from spinach are different than the others.

3.1 The analysis of certified reference material (CRM)

The method was validated by the analysis of certified reference material (CRM). The quantities of analytes were determined to be compatible with the certificate values and the results of the analysis were proved to be correct. The results are given in the Table 5. High percent recovery results were found to be in the range of 90.0–105.9.

Table 5 Certificate, analysis and recovery values (mg/kg) of Certified Reference Material (CRM- NCSZC85006)

No	Element	Certificate value	Analysis value	Recovery %
1	Sodium (Na)	–	–	–
2	Magnesium (Mg)	7360	7135	96.9
3	Phosphorus (P)	5300	5416	102.2
4	Potassium (K)	5790	5668	97.9
5	Calcium (Ca)	53,100	55,020	103.6
6	Aluminum (Al)	2950	3046	103.3
7	Iron (Fe)	1380	1462	105.9
8	Cupper (Cu)	21.10	20.52	97.3
9	Zinc (Zn)	36.20	35.38	97.7
10	Tin (Sn)	–	–	–
11	Arsenic (As)	1.050	0.980	93.3
12	Selenium (Se)	0.080	0.072	90.0
13	Cadmium (Cd)	0.820	0.760	92.7
14	Mercury (Hg)	0.140	0.127	90.7
15	Lead (Pb)	4.970	4.830	97.2

3.2 Statistical analysis

ANOVA's "Independent Sample One-Way Variance Analysis" method was applied by using SPSS 16.0 program on the obtained analysis results. Thus, the results were evaluated statistically according to the sample types (spinach, lettuce or parsley) or where they were taken (Bilecik, Çanakkale etc.). The selected statistical significance level is $p < 0.05$ (95%).

Samples taken from different locations (spinach, lettuce and parsley) were evaluated according to the Independent Sample One Way Variance Analysis (ANOVA) test. As a result, statistically significant differences were found, as can be seen in the Tables 6, 7, and 8.

When evaluated in terms of the averages of the analysis;

- The order of Na, P, Ca, and Zn elements in the samples is parsley > spinach > lettuce
- The order of Mg, K, Al, Fe, Cu, Sn, As, Se and Pb elements in the samples is spinach > parsley > lettuce
- The order of presence in the Cd element is spinach > lettuce > parsley

When all the data were evaluated according to the ANOVA test, it was determined that there was a significant difference between the groups regarding all the elements in terms of their locations.

Table 6 Evaluation of the amount of Na, Mg, P, K and Ca elements according to sample types by "independent sample one-way analysis of variance" ($p < 0.05$)

Sample no	Element	Compared sample type	Significance value	Significance
1	Sodium (Na)	Spinach–Lettuce	0.040	–
		Spinach–Parsley	0.683	+
		Lettuce–Parsley	0.001	–
2	Magnesium (Mg)	Spinach–Lettuce	0.323	+
		Spinach–Parsley	0.784	+
		Lettuce–Parsley	0.129	+
3	Phosphorus (P)	Spinach–Lettuce	0.119	+
		Spinach–Parsley	0.617	+
		Lettuce–Parsley	0.062	+
4	Potassium (K)	Spinach–Lettuce	0.040	–
		Spinach–Parsley	0.104	+
		Lettuce–Parsley	0.369	+
5	Calcium (Ca)	Spinach–Lettuce	0.000	–
		Spinach–Parsley	0.083	+
		Lettuce–Parsley	0.001	–

Table 7 Evaluation of the amount of Al, Fe, Cu, Zn and Sn elements according to sample types by "independent sample one-way analysis of variance" ($p < 0.05$)

Sample no	Element	Compared sample type	Significance value	Significance
1	Aluminum (Al)	Spinach–Lettuce	0.004	–
		Spinach–Parsley	0.168	+
		Lettuce–Parsley	0.002	–
2	Iron (Fe)	Spinach–Lettuce	0.000	–
		Spinach–Parsley	0.002	–
		Lettuce–Parsley	0.000	–
3	Cupper (Cu)	Spinach–Lettuce	0.026	–
		Spinach–Parsley	0.032	–
		Lettuce–Parsley	0.032	–
4	Zinc (Zn)	Spinach–Lettuce	0.911	+
		Spinach–Parsley	0.090	+
		Lettuce–Parsley	0.086	+
5	Tin (Sn)	Spinach–Lettuce	0.020	–
		Spinach–Parsley	0.020	–
		Lettuce–Parsley	0.000	–

Table 8 Evaluation of the amount of As, Se, Cd, Hg and Pb and Sn elements according to sample types by "independent sample one-way analysis of variance" ($p < 0.05$)

Sample no	Element	Compared sample type	Significance value	Significance
1	Arsenic (As)	Spinach–Lettuce	0.002	–
		Spinach–Parsley	0.044	–
		Lettuce–Parsley	0.098	+
2	Selenium (Se)	Spinach–Lettuce	0.014	–
		Spinach–Parsley	0.003	–
		Lettuce–Parsley	0.716	+
3	Cadmium (Cd)	Spinach–Lettuce	0.000	–
		Spinach–Parsley	0.000	–
		Lettuce–Parsley	0.004	–
4	Mercury (Hg)	Spinach–Lettuce	0.000	–
		Spinach–Parsley	0.002	–
		Lettuce–Parsley	0.002	–
5	Lead (Pb)	Spinach–Lettuce	0.000	–
		Spinach–Parsley	0.002	–
		Lettuce–Parsley	0.076	+

4 Conclusions

Pollution levels in soil are constantly increasing due to poor agricultural practices, mining, industrial activities and disposal of urban waste. In addition, soil is polluted through fertilizers and pesticides used in agriculture. As a result, heavy metals are contaminated into the food production chain. Heavy metals are not biodegradable and therefore build up in the important organs of the people's body.

The samples were obtained from food companies and markets selling in the Marmara Region. The collection of the samples was carried out in the winter months because, according to the literature, the season when the heavy metals can accumulate in the greatest amount is the winter months. The other important reasons for this are the increase in population and industry in those regions.

It was determined that the amounts of analytes in the certified reference substance used were in accordance with the certificate values and the results of the analyzes were proved to be correct. The results of As, Cu, Zn, Cd and Pb investigated in the examples are generally close to previous studies. Significant differences were observed in the amount of metal in samples taken from the same variety and from different places.

Heavy metals pose danger and risk in human and all living life as global pollution factors. Depending on the factors such as dose exposure, genetics, immune resistance and general health, age, and nutritional level, they cause various diseases, primarily cancer.

Therefore, starting from production; It is necessary to investigate the ways of contamination and prevention of all kinds of heavy metals that are contaminated with the environment at stages such as storage, packaging, preservation and consumption and that may be harmful to human health.

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Declarations

Competing interest The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this published article.

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