Evaluation of cadmium, lead, copper, iron and zinc in Turkish dietary vegetable oils and olives using electrothermal and flame atomic absorption spectrometry

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RESUMEN

Evaluación de cadmio, plomo, cobre, hierro y zinc en aceites vegetales y aceitunas de la dieta turca mediante espectrometría electrotérmica y absorción atómica de llama.

Se han determinado los metales Cd, Cu, Pb, Fe y Zn en aceites vegetales comestibles (soja, girasol, flores, nueces, maíz y aceite de oliva) y aceitunas (aceitunas-1, negra, verde, negra machacadas con semillas y verde machacadas con semillas) mediante espectrometría de absorción atómica electrotérmica (ETAAS) utilizando como modificador químico la mezcla Sc + Ir + NH$_4$H$_2$PO$_4$ y mediante espectrometría de absorción atómica de llama (FAAS) tras digestión con microondas. Se estudiaron las temperaturas de pirólisis y atomización para Cd, Pb y Cu en disolución con y sin modificador. Los límites de detección (LOD) fueron: 0.1, 0.6, 0.9, 15.0 y 12.0 mg L^{-1} para Cd, Cu, Pb, Fe y Zn, respectivamente. La precisión del procedimiento propuesto se confirmó mediante el análisis de materiales estándar de referencia (SRM) 1577b de hígado de bovino y mediante una solución de muestra sembrada. Los resultados se compararon con los valores certificados. Las desviaciones estándar fueron inferiores al 7% y el rango de las recuperaciones obtenidas de 96 a 101%. La mezcla propuesta Sc + Ir + NH$_4$H$_2$PO$_4$ se aplica a la determinación de Cd, Pb y Cu en los aceites y en aceitunas. Los resultados encontrados en las muestras se compararon con los recogidos en las reglamentaciones internacionales y nacionales de calidad de los alimentos, así como con los valores encontrados en la bibliografía.

PALABRAS CLAVE: Aceites y aceitunas – ETAAS y Faas – Metales – Modificador Químico.

SUMMARY

Evaluation of cadmium, lead, copper, iron and zinc determinations in Turkish dietary vegetable oils and olives using electrothermal and flame atomic absorption spectrometry

The Cd, Pb, Cu, Fe and Zn contents of some edible vegetable oils (soybean, sunflower, flower, nut, corn and olive) and of olives (olive-1, black, green, black crushed with seeds and green crushed with seeds) were determined and evaluated by an electrothermal atomic absorption spectrometer (ETAAS) using an Sc + Ir + NH$_4$H$_2$PO$_4$ chemical modifier mixture and flame atomic absorption spectrometer (FAAS) after microwave digestion. The pyrolysis and atomization temperatures of Cd, Pb and Cu in sample solutions with and without the modifier mixture were investigated. The limits of detection (LOD) for analytes found are 0.1, 0.6, 0.9, 15.0 and 12.0 µg L^{-1} for Cd, Cu, Pb, Fe and Zn, respectively. The accuracy of the procedure proposed was confirmed by analyzing bovine liver 1577b standard reference material (SRM) and a spiked sample solution. The results of the analytes found were compared with certified and added values. The relative standard deviations of the analytes found were lower than 7% and the percent of recoveries obtained ranges from 96 to 101%. The Sc + Ir + NH$_4$H$_2$PO$_4$ mixture proposed was applied for the determination of Cd, Pb and Cu in oils and olives. The results of analytes found in the samples were compared with international and national food quality guidelines as well as with literature values.

KEY-WORDS: Chemical modifier – ETAAS and FAAS – Metals – Oils and olives.

1. INTRODUCTION

The contents of Cd, Cu, Pb, Fe and Zn in edible vegetable oils (soybean, sunflower, flower, nut, corn and olive) and in olive samples are very important for their toxicological effects on human nutrition and health because of the large amounts of these samples consumed by people as food in their daily diets (Bakkali *et al.*, 2009; Benincasa *et al.,* 2007; Mendil *et al.*, 2009; Roca *et al.*, 2000; Sahan *et al.*, 2007). Dietary vegetable oils and olives play an essential role in human nutrition and during digestion and metabolism they may react with some chemicals, in particular with oxygen (Mendil *et al.*, 2009). While iron and copper are regulated as criteria for the qualities of edible olive oils and olives, lead and cadmium are considered as real pollutants reported in legislations and literature (IOOC, 2003, Bakkali *et al.*, 2009; Economic European Communities, 2006). Cu, Fe and Zn are important for human health as essential nutrients having different biological functions in all living organisms such as increasing oil oxidation, while Cd and Pb are very important for humans due to their toxicities and metabolic roles even at low concentrations (Mendil *et al.*, 2009; Zhu *et al.*, 2011; Ansari *et al.*, 2009; Guldas, 2008; Nunes *et al.*, 2011; Sardans *et al.*, 2010). The sources of Cd, Cu, Pb, Fe and Zn contamination in these samples might come from the soil as natural metal sources, manufacturing, packaging process and environmental pollution (Mendil *et al.*, 2009; Zeiner *et al.*, 2005; Benincasa *et al.*, 2007). It is important to determine the levels

of these metals in edible oils and olives and to point out the possible contamination levels of the analytes for human health.

The analytical techniques commonly used for the determination of analytes in vegetable oils and biological samples are the inductively coupled plasma optical emission spectrometry (ICP-OES), inductively coupled plasma mass spectrometry (ICP-MS) (Sahan *et al.*, 2007; Zeiner *et al.*, 2005; Souza *et al.*, 2008), electrothermal atomic absorption spectrometry (ETAAS) and flame atomic absorption spectrometry (FAAS). Among these techniques, ETAAS and FAAS are extensively employed for the determination of the total trace element contents in samples because of their high sensitivities, lower detection limits and low costs (Mendil *et al.*, 2009; Sardans *et al.*, 2010; Nunes *et al.*, 2011). However, in general, the determination of metals in oil samples by atomic spectrometric methods is highly difficult due to high background signals obtained from organic contents of the sample matrix and the volatilization of analytes together with organic compounds (Angioni *et al.*, 2006; Sahan *et al.*, 2007; Souza *et al.*, 2008; Nunes *et al.*, 2011). Some sample pretreatment methods such as extraction, pre-concentration and dilution are required to minimize the organic matrices in vegetable oils before using a spectroscopic technique. In some cases, these methods are tedious and time consuming with the consequent risk of sample contamination, analyte loss and an incomplete dissolution of the sample matrix (Carrilho *et al.*, 2002; Ansari *et al.*, 2009). Microwave digestion systems, used for laboratory applications, offer enormous advantages in dissolving the samples (Angioni *et al.,* 2006; Sahan *et al.*, 2007; Souza *et al.*, 2008; Ansari *et al.*, 2009; Nunes *et al.*, 2011). Platform atomization, matrix modification, integrated absorbance and powerful background correction techniques together with ETAAS have been used to overcome these problems. Different permanent modifiers (W-Rh, W-Ir and W-Ru) (Lima *et al.*, 2002a, b) and other suitable modifiers, such as Sc + Pd + NH_4NO_3 (Acar, 2005), Sc + Ru + $(NH_4)_2HPO_4$ (Acar, 2004) and organic palladium and palladium - magnesium chemical modifiers (Kowalewska *et al.*, 1999) have been used for the determination of Cd, Cu and Pb in various samples in order to stabilize the analytes to higher permissible pyrolysis temperatures and to reduce such interferences in a sample matrix prior to the atomization steps. An Sc + Ir + $NH_4H_2PO_4$ modifier mixture has not been used in previous studies.

The aims of the present study were to determine the contents of Cd, Pb, Cu, Fe and Zn in dietary vegetable oils and olives produced in Turkey by ETAAS using Sc + Ir + $NH_4H_2PO_4$ and FAAS after a microwave digestion process and to compare the results of the analytes found in the samples with national and international legislations (IOOC, 2003, Economic European Communities, 2006) as well as values found in the literature with regard to potential human health risks.

2. MATERIALS AND METHODS

2.1. Reagents

All reagents and acids used were of analytical reagent grade unless otherwise stated. All solutions were prepared by dissolving Nitric acid (65% m/m), Triton X-100 (99.9% m/m) and H_2O_2 (30% m/m) (Merck, Darmstadt, Germany) in ultra - pure water (resistivity 18 MΩ cm^{-1}) obtained from ultra - pure water system (Nanopure Infinity, Barnstead, P/N-1161, Dubuque, USA). All solutions were stored in high density polypropylene bottles. All glassware and plastic materials and auto sampler cups were cleaned by soaking in an HNO_3 (20% v/v) solution for 24 h, then rinsed four times with ultra - pure water and dried. An auto sampler washing solution containing HNO_3 (0.1% v/v) and Triton X-100 (0.1% v/v) was used to prevent clogging of the auto sampler pipette and to improve the dispersion of the sample solution onto the platform (Lima *et al.*, 2002a, b).

A Scandium (III) stock solution (4.00 g L^{-1}) was prepared by dissolving 310 mg Sc_2O_3 (Merck, 99.99% pure) in 2 mL of concentrated HNO_3 by heating on a hot plate and diluting to 50 mL. Stock standard solution of Ir (2.00 g L^{-1}) was prepared by dissolving 235 mg IrO_2 (Merck) in 3 mL HNO_3 (10% v/v) and diluted to 100 mL. The $NH_4H_2PO_4$ solution (4% m/v) was prepared by dissolving 4.00 g $NH_4H_2PO_4$ (Merck, 99.99% pure) in a sufficient volume of ultra - pure water, transferring it to a 100-mL volumetric flask and diluting to the mark with ultra - pure water. All modifier solutions were diluted as required.

The element standard solutions used for calibration were freshly prepared by diluting stock standard solutions of Cd, Cu, Pb, Fe and Zn (1000 mg L^{-1} of each) obtained from BDH chemicals (Poole, UK) in a nitric acid solution (0.1% v/v) immediately before use.

2.2. Sample collection

Fifty-three oil samples (8 soybean, 12 sunflower, 8 flower, 8 nut, 8 corn and 9 olive oil) along with 70 olive samples (20 olive-1, 15 black, 12 green, 10 black crushed and 13 green crushed samples) coming from various plants and regions of Turkey were obtained from different supermarkets in Ankara on different dates. The seeds of some of black and green olive samples were separated by hand or crushed and the flesh was homogenized with a stainless blender in order to assess possible contamination from the olive seed. The samples were weighed and transferred into poly-ethylene storage containers for analysis. Bovine liver 1577b standard reference material (SRM) purchased from the National Institution of Standards and Technology (NIST) was used for recovery tests.

2.3. Instrumentation

A Hitachi (Japan) Model 180/78 graphite furnace and 180/80 flame atomic absorption

spectrometer equipped with a Zeeman effected background corrector and an automatic data processing unit (180/205) was used for all the absorption measurements of Cd, Cu, Pb, Fe and Zn. Hitachi pyrolytically coated graphite tubes (P/N-190/6007) inserted with graphite platforms (P/N-190/6008) and integrated (peak area) mode were used for atomization throughout the study. Single element hollow cathode lamps of Cd (10 mA and 228.8 nm), Cu (10 mA and 324.8 nm), Pb (10 mA and 283.3 nm), Fe (12.5 mA and 248.3 nm) and Zn (10 mA and 213.9 nm) were used as radiation sources. The slit width was 1.3 nm for all lamps. The instrumental parameters and operating conditions of ETAAS and FAAS for the analytes recommended by the manufacturer were used, unless otherwise stated. Argon (99.98%, v/v) was used as a carrier gas during all stages except for atomization in ETAAS. The graphite furnace temperature program optimized for the determination of Cd, Cu and Pb by ETAAS with $Sc + Ir + NH_4H_2PO_4$ modifier mixture solution is given in Table 1. A twenty µL volume of calibration or sample solution together with modifier solution was injected into the platforms by an autosampler (P/N-170/126). An air-acetylene flame was used for the determinations of Fe and Zn by FAAS. A Milestone Ethos microwave oven (MLS Ethos 1600, Italy), equipped with temperature and pressure sensor, Teflon digestion bombs and vessels was used to dissolve the samples.

2.4. Digestion of samples

A microwave-assisted digestion procedure was used for the decomposition of samples in a shorter time. Triplicate samples of oils (1.00 - 2.00 g portion of each oil), olives (1.00 - 2.00 g portion of each olive) and bovine liver 1577b (0.50 - 1.00 g) were accurately weighed into separate Teflon digestion vessels. Two mL of ultra-pure water, 6 mL of HNO_3 (65% m/m) and 2 mL of H_2O_2 (30% m/m) were added into each vessel (Mendil et al., 2009; Bakkali et al., 2009), and kept at laboratory temperature for 4 h with a gentle swirl of the acid mixture to dissolve the sample without heating. The vessel was closed, placed inside the microwave digestion system and digestion was carried out at 160 °C at maximum

power (800W) for 20 - 25 min to decompose the organic matter (Lima et al., 2002a). After cooling, the digestion vessel was opened, placed on a hot plate and heated to 3 mL to evaporate excess acid. The resulting solution was transferred into a 25-mL volumetric flask after washing the interior surface of the digestion vessel with nitric acid (1% v/v) three times and diluting the solution with ultra - pure water containing nitric acid (1% v/v) and then kept as a stock sample solution. Blank solutions were also prepared by using the digestion procedure given above to check the possible analyte contaminations in the reagents used for the sample preparation. Three portions of an oil sample (1.00 g of each) were accurately weighed into three Teflon digestion vessels and known amounts of calibration standard solutions were added into the second and third digestion vessels without any addition to the first vessel. The digestion procedure was repeated to check the validity and efficiency of the microwave-assisted digestion using the standard addition method (Ansari et al., 2009).

2.5. Optimization studies of analytes for ETAAS determinations

The use of a chemical modifier is required for the determinations of Cd, Pb and Cu in vegetable oils and olive samples to reduce background absorbance coming from a sample matrix such as organic and inorganic components. For this purpose, 2.0 g L^{-1} Sc; 2.0 g L^{-1} Sc + 0.4 g L^{-1} Ir and 2.0 g L^{-1} Sc + 0.4 g L^{-1} Ir + 5.0 g L^{-1} $NH_4H_2PO_4$ modifier systems (Acar 2004; Acar, 2005) were tested. Some of the sample solutions were diluted to the desired concentrations (0.05 < absorbance unit) with ultra - pure water containing a nitric acid (0.1% v/v) and Triton X-100 (0.2% v/v) mixture to find optimum parameters for the ETAAS determinations. One milliliter of a modifier solution was mixed with 1 mL of a sample solution and 20 µL of this mixture were injected onto the platform. Drying, pyrolysis, atomization and cleaning temperatures, ramp and hold times in the graphite atomizer were modified in order to obtain maximum absorbance values for the analytes. The optimized heating temperature program proposed for the determination of analytes by ETAAS with an $Sc + Ir + NH_4H_2PO_4$ mixture is given in Table 1.

Table 1
Heating program for the determinations of Cd, Cu and Pb in samples by ETAAS with $Sc + Ir + NH_4H_2PO_4$ modifier mixture

Step	Temperature (°C)			Time (s)		Ar flow rate (mL min⁻¹)
	Cd	Cu	Pb	Ramp	Hold	
Dry-1		50-130		20	–	250
Dry-2		130-300		15	15	250
Pyrolysis	900	1,300	1,250	20	20	250
Atomization	1,500	2,700	2,000	0	5	0
Cleaning	2,650	2,800	2,650	0	3	250

Pyrolysis and atomization curves for Cd, Pb and Cu in an olive sample with and without Sc, Sc + Ir and Sc + Ir + $NH_4H_2PO_4$ modifier solutions were performed and are shown in Figure 1 for Cd, Pb and Cu. The mean values of three absorbance measurements for the analytes obtained from Sc, Sc + Ir and Sc + Ir + $NH_4H_2PO_4$ chemical modifiers are plotted.

3. RESULTS AND DISCUSSION

3.1. Thermal stabilization studies of Cd, Cu and Pb with modifiers

The purpose of using the chemical modifier is to provide high thermal stabilization of the analytes by increasing pyrolysis temperatures up to higher
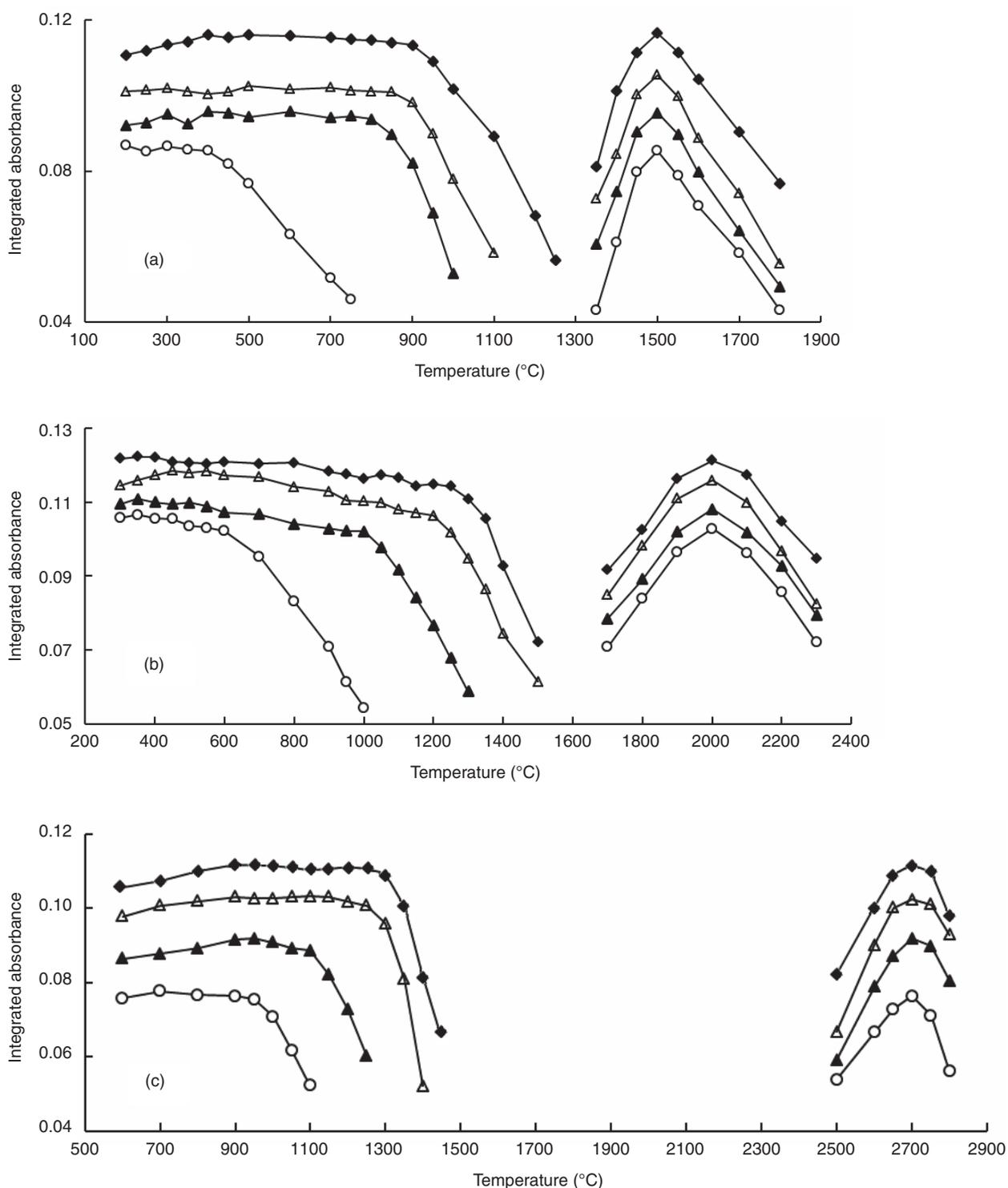

Figure 1
Pyrolysis and atomization curves for (a) Cd, (b) Pb and (c) Cu in an olive sample solution (Dilution ratio is 2 for Cd, Pb and Cu) without (O) and with 20 μg Sc (▲), 20 μg Sc + 4 μg Ir (Δ) and 20 μg Sc + 4 μg Ir + 50 μg $NH_4H_2PO_4$ (♦).

permissible values without a loss in analytes and to reduce interferences coming from the sample matrix. Thermal stabilization studies of Cd, Pb and Cu in an olive sample solution with and without the Sc, Sc + Ir and Sc + Ir + $NH_4H_2PO_4$ modifier mixture were performed by ETAAS. The addition of $NH_4H_2PO_4$, NO_3^- and Triton X-100 to the samples and calibration solutions may reduce the Sc (III) and Ir (IV) ions along with the analytes into their metallic forms to obtain an analyte-modifier interaction and to increase the thermal stability of the analyte with the modifier (Byrne et al., 1993; Tsalev et al., 2002). The maximum absorbance values and pyrolysis temperatures of analytes in the sample solution were obtained with the Sc + Ir + + $NH_4H_2PO_4$ (Fig.1) and the pyrolysis temperatures of the analytes found by the Sc + Ir + $NH_4H_2PO_4$ modifier mixture are also given in Table 1. As can be seen, the pyrolysis temperatures of the analytes are sufficient to remove interferences in the sample matrix without the risk of analyte loss. The pyrolysis temperatures of the analytes obtained with Sc + Ir + $NH_4H_2PO_4$ were compared with different modifier mixtures given in the previous studies (Slaveykova and Tsalev, 1990; Acar, 2005) and similar results were observed. The atomization temperatures of the analytes from the atomization curves (Fig. 1) are also given in Table 1. Therefore, the Sc + Ir + $NH_4H_2PO_4$ modifier mixture proposed was used for the determinations of Cd, Pb and Cu by ETAAS in subsequent experiments.

3.2. Analytical characteristics

The concentrations of trace metals in the sample solutions were determined using the calibration graph method. The calibration graphs against aqueous standard solutions of analytes were linear and the dynamic ranges obtained were 0.3 - 6 µg L^{-1} for Cd, 3.0 - 80 µg L^{-1} for Pb, 2.0 - 80 µg L^{-1} for Cu, 0.05 - 6.0 mg L^{-1} for Fe and 0.04 - 1.0 mg L^{-1} for Zn. The correlation coefficients (R^2) of all calibration graphs for the analytes studied were higher than 0.99. The lower limits of dynamic range were considered as limits of quantification (LOQ) values for the analytes found. LOQ (10 σ_b/m, where, σ_b is the standard deviation of the blank measurements, m is the slope of the calibration curve), LOD (3 σ_b/m) and characteristic mass (m_o, the mass of analyte related to 0.0044 absorbance unit) values for Cd, Pb and Cu, based on integrated absorbance with Sc + Ir + + $NH_4H_2PO_4$, were calculated from 20 consecutive measurements of blank solutions according to IUPAC (Barrera et al., 1993; Commission on Spectrochemical and Other Optical Procedures for Analysis, 1978). The m_o values of Cd, Pb and Cu found were 0.8, 8.3 and 19 pg, respectively. These m_o values of Cd, Pb and Cu were compared with the results obtained from previous studies (Lima et al., 2002b; Tamasi and Cini, 2004; Zhu et al., 2011; Saçmacı and Kartal, 2011) and similar values were confirmed.

3.3. Recovery studies

The validity of the proposed method for the determinations of Cd, Pb and Cu by ETAAS using the Sc + Ir + $NH_4H_2PO_4$ mixture was tested by analyzing bovine liver 1577b (SRM). Recovery tests for analytes in an oil sample solution were supported by the standard addition method. Iron and Zn were also determined in these samples by FAAS. The results of the analytes found are given in Table 2. As seen in Table 2, the recovery values were between 96% and 101% and they were in good agreement with the certified and added values. The relative standard deviations of analytes found were below 7% (range is from 3.4 to 6.2%). Based on the results found, it was concluded that the Sc + Ir + $NH_4H_2PO_4$ modifier mixture could be applied for the determinations of Cd, Pb and Cu in SRM, oils and olive samples with lower detection limits and high accuracy. The microwave digestion system was also controlled with respect to the recovery values and it was preferred for digesting such samples.

3.4. Determination of analytes in oils and olive samples

Cadmium, Pb, Cu, Fe and Zn in vegetable oils and olive samples by ETAAS using Sc + Ir + + $NH_4H_2PO_4$ and FAAS were determined. Three samples for each oil and olive were acquired. Analytes in a sample solution were analyzed individually in five replicate measurements with a 95% confidence level. For each analyte, the average ± confidence interval was obtained. The results of analytes found in samples are reported as the mean ± standard deviation (S.D.) (min. - max. value) obtained from the average ± confidence intervals. The concentration levels of Cd, Pb, Cu, Fe and Zn found in samples are given in Table 3. All metal concentrations found in samples were on a wet weight as mg kg^{-1} except cadmium. The minimum, mean and maximum values of analytes found are shown in Figure 2 and the differences in the results of analytes obtained from oils and olive samples are clearly depicted. The sources of metals in vegetable oils and olive samples may be attributed to multiple factors, such as soils and fertilizers used for cultivation (Sahan, 2011). Processing equipment, storage and packaging containers, etc. are also important sources for tinted metal contamination in food. In addition, it has been reported that olives treated with chemicals contain more Pb than naturally processed olives (Sahan, 2011).

Cadmium is known as a highly toxic trace metal and excessive consumption of it may give rise to human diseases such as renal, pulmonary, hepatic, skeletal and cancer (Zhu, et al., 2011). The cadmium concentration levels in oil and olive samples reported in the literature are given in Table 4. As can be seen in Table 4 and Figure 3, the minimum and maximum values of Cd found in oil

Table 2
Recovery tests for analytes in bovine liver 1577b (SRM) and an oil sample solution

Element	Certified /Added[a]	Found[b]	Recovery (%)
Bovine liver 1577b	**Concentrations, mg kg^{-1}**		
Cd	0.50 ± 0.03	0.48 ± 0.02	96
Cu	160 ± 8	158 ± 7	99
Pb	0.129 ± 0.004	0.13 ± 0.01	101
Fe	184 ± 15	181 ± 9	98
Zn	127 ± 16	124 ± 6	98
Oil sample solution	**Concentrations, µg L^{-1}**		
Cd	0	1.44 ± 0.06	0
	1.0	2.40 ± 0.08	98
	2.0	3.41 ± 0.13	99
Cu	0	5.4 ± 0.2	0
	10	14.8 ± 0.5	96
	20	24.9 ± 1.0	98
Pb	0	6.2 ± 0.3	0
	10	15.7 ± 0.7	97
	20	25.9 ± 1.2	99
Fe	0	124 ± 6	0
	200	312 ± 14	96
	400	528 ± 23	101
Zn	0	70 ± 3	0
	200	262 ± 10	97
	400	461 ± 19	98

[a] Added aqueous standard solutions for an oil sample solution; [b] Mean of five replicate measurements for an analyte with 95% confidence level, $\bar{x} \pm \frac{ts}{\sqrt{N}}$.

the samples in the present study are higher than the literature values, but they are in agreement with the maximum contamination levels of Cd reported in the legislation (Economic European Communities, 2006). The concentration ranges of cadmium found in olives are in agreement with the literature values. On average, the Cd levels found in the Turkish dietary oils and olives are lower or about 100 µg kg^{-1} Cd for vegetable samples given in the Turkish food codex regulations (TKB, 2002).

The maximum concentration levels of Pb found in the oils (Table 3 and Figure 2) are higher than the literature values (Table 4), but they are lower or about 0.1 mg kg^{-1} for vegetable and refined oils given in the Turkish standards (TS 2812, 1991; TS 3606, 1997), Turkish food codex regulations (TKB, 2002) and the legislations (IOOC, 2003; Economic European Communities, 2006). The minimum and maximum Pb concentration levels found in the olives (Table 3 and Figure 2) are in agreement with the literature values given in Table 4. Lead and Cd are similar to each other with no beneficial effects on human metabolism and producing toxicity. Some

health disorders from Pb intake are sleeplessness, tiredness, hearing and weight loss (Sahan et al., 2007).

The minimum and maximum values of Cu found in the samples (Table 3, Table 4 and Figure 2) are in agreement with some of the literature values given in Table 4. The safe upper level of Cu allowed is 10 mg for a 60 kg adult (EVM, 2003).

Fe values found in the samples (Table 3 and Figure 2) are in agreement with some of the literature values (Table 4). The maximum concentration of Fe allowed is 150 mg kg^{-1} (Biricik and Basoğlu, 2006). Iron deficiency is frequently related to anemia and it reduces working capacity and impairs intellectual development (Schümann et al., 2007). High concentrations of iron may lead to tissue damage and the formation of free radicals (Biricik and Basoğlu, 2006; Lopez et al., 2008; Sahan et al., 2007).

The zinc levels found in the oils and olives (Table 3 and Figure 2) are in agreement with some of the literature values (Table 4). Zinc is known to be an essential element for metabolic functions in the human body and deficiency of it can lead to
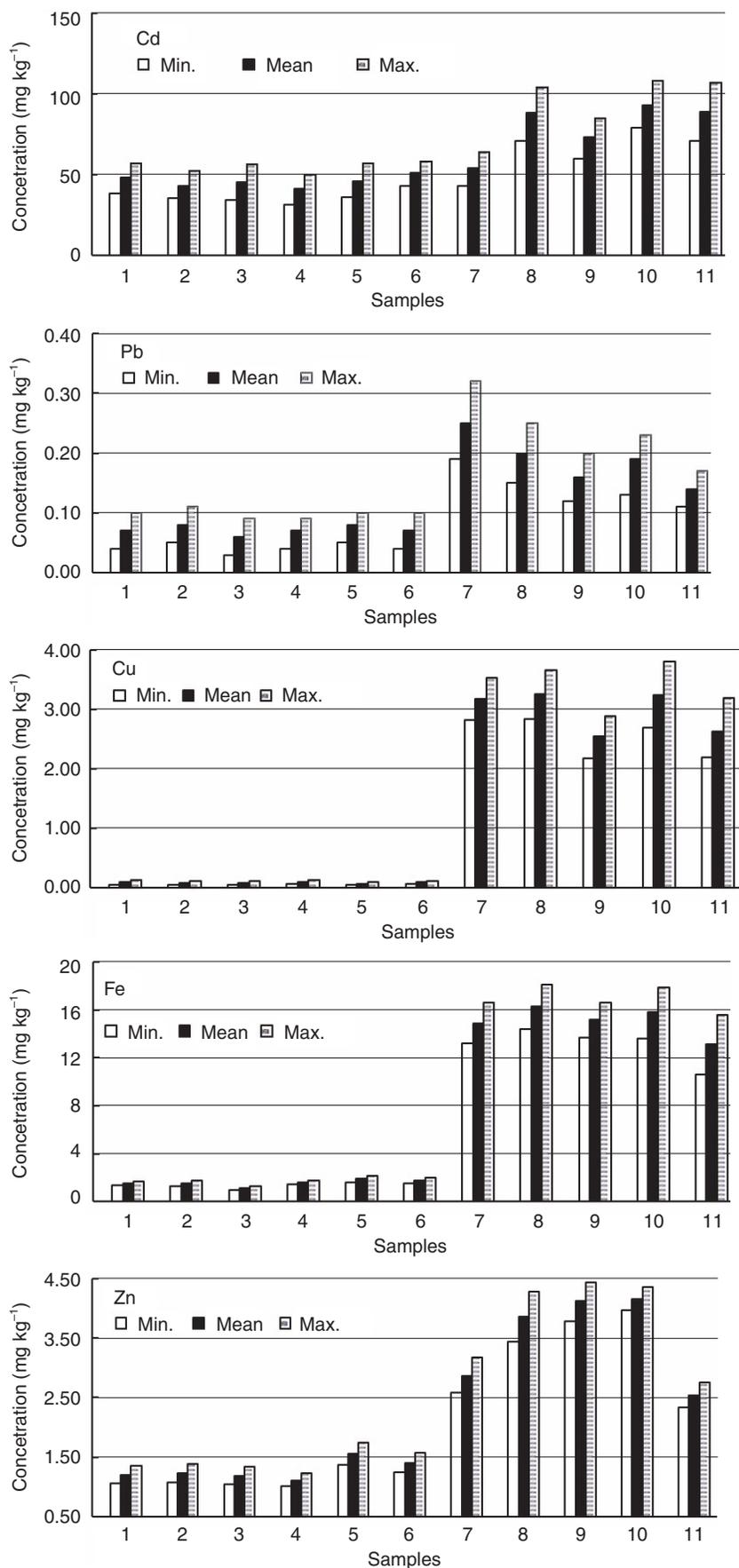
Figure 2
Distributions of Cd, Pb, Cu, Fe and Zn levels in oils and olives. 1, soybean oil; 2, sunflower oil;
3, flower oil; 4, nut oil; 5, corn oil; 6, olive oil; 7, olive-1 sample; 8, olive black; 9, olive green;
10, olive black crashed sample; 11, olive green crashed sample.

Table 3
Results of analytes found in edible vegetable oils and olive samples

Sample	Number	Concentrations[a] (mg kg^{-1}, $\bar{x}$ ± S.D., Min. - Max. value)				
		Cd[b]	Pb	Cu	Fe	Zn
Oil samples						
Soybean	8	48 ± 7	0.07 ± 0.02	0.09 ± 0.02	1.52 ± 0.14	1.21 ± 0.11
		38 – 57	0.04 – 0.10	0.05 – 0.13	1.33 – 1.68	1.07 – 1.36
Sunflower	12	43 ± 6	0.08 ± 0.01	0.07 ± 0.01	1.51 ± 0.18	1.23 ± 0.12
		35 – 52	0.05 – 0.11	0.04 – 0.10	1.30 – 1.71	1.08 – 1.39
Flower	8	45 ± 8	0.06 ± 0.01	0.08 ± 0.01	1.13 ± 0.13	1.19 ± 0.10
		34 – 56	0.03 – 0.09	0.04 – 0.11	0.96 – 1.29	1.05 – 1.34
Nut	8	41 ± 7	0.07 ± 0.01	0.09 ± 0.02	1.60 ± 0.14	1.11 ± 0.08
		31 – 50	0.04 – 0.09	0.06 – 0.13	1.41 – 1.78	1.01 – 1.24
Corn	8	46 ± 8	0.08 ± 0.01	0.06 ± 0.01	1.87 ± 0.22	1.56 ± 0.15
		36 – 57	0.05 – 0.10	0.04 – 0.09	1.59 – 2.14	1.38 – 1.74
Olive	9	51 ± 5	0.07 ± 0.02	0.09 ± 0.01	1.76 ± 0.19	1.41 ± 0.12
		43 – 58	0.04 – 0.10	0.06 – 0.11	1.53 – 1.99	1.25 – 1.58
Olive samples						
Olive-1	20	54 ± 8	0.25 ± 0.03	3.18 ± 0.31	14.9 ± 1.3	2.86 ± 0.24
		43 – 64	0.19 – 0.32	2.81 - 3.53	13.2 – 16.6	2.58 – 3.18
Black	15	88 ± 14	0.20 ± 0.03	3.26 ± 0.37	16.3 ± 1.6	3.86 ± 0.38
		71 – 104	0.15 – 0.25	2.83 – 3.66	14.4 – 18.1	3.44 – 4.28
Green	12	73 ± 10	0.16 ± 0.02	2.55 ± 0.28	15.2 ± 1.2	4.12 ± 0.29
		60 – 85	0.12 – 0.20	2.17 – 2.88	13.7 – 16.6	3.78 – 4.44
Black crushed with seeds	10	93 ± 12	0.19 ± 0.02	3.24 ± 0.51	15.8 ± 1.9	4.16 ± 0.16
		79 – 108	0.13 – 0.23	2.69 – 3.80	13.6 – 17.9	3.96 – 4.36
Green crushed with seeds	13	89 ± 14	0.14 ± 0.01	2.62 ± 0.37	13.1 ± 2.1	2.54 ± 0.18
		71 – 107	0.11 – 0.17	2.19 – 3.09	10.6 – 15.6	2.33 – 2.75

[a] Mean of five replicate measurements for an analyte with 95% confidence level, $\bar{x} \pm \dfrac{ts}{\sqrt{N}}$.
[b] µg kg^{-1}

loss of appetite, growth retardation, skin changes and immunological abnormalities (Mendil et al., 2009; Zhu, et al., 2011).

The recommended daily intake values of Zn and Cu are 15 mg Zn for adult males, 12 mg Zn for adult females and 1.5-3.0 mg Cu for an adult person (Mendil et al., 2009). For an average adult (60 kg body weight), the provisional tolerable daily intake (PTDI) for Fe, Cu, Pb and Zn are 48 mg, 3 mg, 214 µg and 60 mg, respectively (Joint FAO/WHO, 1999). The maximum permissible values of analytes given in Turkish standards (TS 2812, 1991; TS 3606, 1997) and Turkish food codex regulations (TKB, 2002) for vegetable oils and refined liquid oils are 1.5, 0.1, 0.1 mg kg^{-1} for Fe, Cu and Pb, respectively. The maximum permissible values of Cu and Fe for olive oil and non-refined vegetable oil given in Turkish food codex regulations (TKB, 2002) are 0.1 and 0.4 mg kg^{-1} for Cu and 3.0 and 5.0 mg kg^{-1} for Fe, respectively. The approved contents of some metals in oils from national and international requirements are 0.05 mg kg^{-1} Cd, 1 - 1.5 mg kg^{-1} Fe, 0.1 mg kg^{-1} Cu and Pb, respectively (Kowalewska et al., 2005; Zhu et al., 2011). The maximum recommended levels of Fe, Cu, Pb and Cd reported

in the legislation (IOOC, 2003; Economic European Communities, 2006) are 0.3 mg kg^{-1} for Fe, 0.1 mg kg^{-1} Cu and Pb for oils and olives, 0.2 mg kg^{-1} Cd for soybeans and 0.050 mg kg^{-1} Cd for vegetables, pine nuts, etc., respectively. The highest concentration levels of analytes found in the olive samples (Table 3 and Figure 2) are lower than the maximum values reported in the literature (Sahan and Basoğlu, 2004). The various metal levels are below the safe limits specified for table olives by Turkish local food standards (TS 774, 2003).

4. CONCLUSIONS

It has been demonstrated that digestion treatment of oils and olive samples with a microwave oven using nitric acid and hydrogen peroxide mixture was fast and simple. The determinations of Cd, Pb and Cu in the samples by ETAAS using the Sc + Ir + + NH$_4$H$_2$PO$_4$ modifier mixture proposed, and Fe and Zn by FAAS have been performed with low detection limits, acceptable sensitivity and selectivity. The use of the Sc + Ir + NH$_4$H$_2$PO$_4$ modifier mixture favors the vaporization of the sample matrix and reduces

Table 4
Comparison of analyte results found in oil and olive samples with literature values
(Minimum - Maximum values given in mg kg^{-1})

Element	Sample	Literature values	This study
Cd	Oil[a]	0.09 - 4.57 (Mendil *et al.*, 2009); 2.64 - 8.43 (Zhu *et al.*, 2011); 0.2 - 4.5 (Pehlivan *et al.*, 2008); 0.0013 - 0.0123 (Bakkali *et al.*, 2009)	31 - 58
	Olive	0.06 - 0.36 in black olive samples (Şahan and Basoğlu, 2004); 0.08 - 0.15 in black and green olives (Şahan, 2011); 3.2 - 8.1 (Zeina *et al.,* 1997); 0.06 (Madejan *et al.*, 2006).	0.04 - 0.11
Pb	Oil	0.009 - 0.018 (Zhu *et al.*, 2011); 0 - 0.0074 (Pehlivan *et al.*, 2008); 0-0.03 (Mendil et al., 2009); 0.074 - 0.104 (Bakkali *et al.*, 2009)	0.04 - 0.11
	Olive	0.79 - 3.68 (Şahan and Basoğlu, 2004); 0.71 in black olives and 0.75 in green olives (Şahan *et al.*, 2007).	0.11 - 0.32
Cu	Oil	0.02 - 0.33 (Garrido *et al.*, 1994); 0.0103 - 0.0850 (Pehlivan *et al.*, 2008); 0 - 130 (Llorent-Martínez et al., 2011a, b); 0 - 0.71 (Mendil et al., 2009); 0.004 - 0.5 (Juranovic Cindric *et al.*, 2007); 0.82 - 4.51 in olive oil (Zeiner *et al.*, 2005); 0.214 - 0.875 (Zhu *et al.*, 2011); 0.019 - 0.096 (Bakkali *et al.*, 2009)	0.04 - 0.13
	Olive	2.85 - 13.01 (Şahan and Basoğlu, 2004); 0.73 - 2.55 in black olives (Şahan *et al.*, 2007); 3.50 - 6.67 in green olives (Biricik and Basoğlu, 2006); 3.99 - 10.93 in directly brined olives (López *et al.*, 2008).	2.17 - 3.80
Fe	Oil	16.2 - 45.3 (Zhu *et al.*, 2011), 0.22 - 220 (Garrido et al., 1994), 52.0 - 291.0 (Mendil *et al.*, 2009), 0.0039 - 0.0352 (Pehlivan *et al.,* 2008), 0 - 0.800 (Llorent-Martínez et al., 2011a, b), 15.3 - 23.3 (Juranovic Cindric et al., 2007); 13.10 - 18.46 (Zeiner *et al.*, 2005); 0.46 - 1.61 (Benincasa *et al.*, 2007)	0.96 - 2.14
	Olive	14.36 - 118.55 (Şahan and Basoğlu, 2004); 3.23 - 15.10 in green table olives (Biricik and Basoğlu, 2006); 3.49 - 7.70 (López *et al.*, 2008).	10.6 - 18.1
Zn	Oil	0.742 - 2.56 (Zhu *et al.,* 2011), 1.03 - 3.08 (Mendil *et al.*,2009), 2.1 - 14.0 (Juranovic Cindric *et al.*, 2007), 0.04 - 0.70 (Garrido *et al.*, 1994), and 0.0184 - 0.2870 (Pehlivan *et al.*, 2008) and 2.82 - 4.03 (Zeiner *et al.*, 2005); 0.009 - 0.037 (Roca *et al.*, 2000)	1.01 - 1.74
	Olive	2.19 - 11.53 (Şahan and Basoğlu, 2004); 8.50 ± 1.74 in black and 10.58 ± 2.01 green types (Şahan *et al.,* 2007).	2.33 - 4.44

[a] in µg kg^{-1} for oil samples.

potential interferences at the atomization stage of analytes. The application of the proposed method for the determination of metals in a certified sample (bovine liver 1577b) and an oil sample has shown that the methodology is accurate. The results obtained for Cd and Pb in oils in the present study may be higher than the literature values, but they are in agreement with the legislation (IOOC, 2003; Economic European Communities, 2006). Generally, the intake of Cd, Pb, Cu, Fe and Zn from daily consumption of oils and olives produced in Turkey may not be a risk for human health and this analytical methodology may also be applicable to analyze similar matrices in food samples.

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