



Anal. Bioanal. Chem. Res., Vol. 6, No. 2, 365-380, December 2019.

Effervescence-assisted Dispersive Liquid-liquid Microextraction for Trace Analysis of Co(II) and Ni(II) from Aqueous Sample Based on Phthalic Acid as a Complexing agent and Co-disperser

Saeed Mohammad Sorouraddin^{a,*}, Mir Ali Farajzadeh^{a,b} and Hossein Najafpour Qarajeh^a

^a*Department of Analytical Chemistry, Faculty of Chemistry, University of Tabriz, Tabriz, Iran*

^b*Engineering Faculty, Near East University, 99138 Nicosia, North Cyprus, Mersin 10, Turkey*

(Received 18 November 2018 Accepted 4 April 2019)

A green effervescence-assisted dispersive liquid-liquid microextraction based on phthalic acid, as a complexing agent and co-disperser, coupled with graphite furnace atomic absorption spectrometry has been developed for the extraction and preconcentration of Co(II) and Ni(II) from aqueous samples. Initially two test tubes were selected. A specified amount of sodium bicarbonate is placed at the bottom of the first conical and dried glass test tube, and then μl -level of 1,1,2,2-tetrachloroethane as an extraction solvent is added. Phthalic acid (as a complexing agent) is added to the aqueous solution placed in the second tube to form phthalate-metal complexes. Then, the content of the second tube is added into the first tube. A reaction between the excess amount of phthalic acid and sodium bicarbonate is immediately occurred, and the produced CO_2 leads to dispersion of the extraction solvent as tiny droplets. In this study, phthalic acid is used simultaneously as a complexing agent and co-disperser. Under the optimum conditions the calibration curves were linear in the ranges of 25.0-1000.0 and 50.0-1000.0 ng l^{-1} for Co(II) and Ni(II), respectively. The detection limits were obtained 9.2 and 15.0 ng l^{-1} for Co(II) and Ni(II), respectively. Extraction recoveries were 99 and 98% for Co(II) and Ni(II), respectively. Enrichment factors were obtained 197.8 and 196.4 for Co(II) and Ni(II), respectively. The relative standard deviations were $\leq 1.3\%$ for intra- ($n = 6$) and ≤ 3.0 for inter-day ($n = 6$) precisions. Finally, the proposed method was successfully applied for the simultaneous analysis of the analytes in environmental water and fruit juice samples.

Abbreviations: DLLME: Dispersive liquid-liquid microextraction, EA-DLLME: Effervescence-assisted- dispersive liquid-liquid microextraction, EF: Enrichment factor, ER: Extraction recovery, GFAAS: Graphite furnace atomic absorption spectrometry, LOD: Limit of detection, LOQ: Limit of quantification, LR: Linear range

Keywords: Cobalt, Nickel, Phthalic acid, Effervescence-assisted dispersive liquid-liquid microextraction, Graphite furnace atomic absorption spectrometry

INTRODUCTION

Heavy metals are widely used in many industries and present relatively low amounts in the environment. They are important pollutants in environment due to their toxic effect on human health. Cobalt and nickel are amongst the most important nonferrous metals. Excessive nickel affects some

organ systems such as the immunological, respiratory, dermal, and cardiovascular ones [1-3]. The maximum admissible limit of nickel in drinking water is 70 $\mu\text{g l}^{-1}$ (WHO, 2008) [4]. Cobalt is a naturally occurring element found in soil, water, rocks, animals and plants. It is used to produce alloys used in industry. It may cause several health problems such as paralysis, diarrhea, low blood pressure, lung irritation, and bone defects [5]. The maximum admissible limit of cobalt in drinking water is not

*Corresponding author. E-mail: ssouredin@tabrizu.ac.ir

mentioned by World Health Organization (WHO, 2008), however, it has been reported $100 \mu\text{g l}^{-1}$ by United States Environmental Protection Agency (USEPA, 2008) [6]. The concentrations of these toxic metals in environmental samples, such as water and food samples must be lower than certain threshold concentrations due to their harmful effects [7]. Therefore, accurate and precise determination of these ions in environmental samples is the major challenge for analytical chemists because of their trace levels. Different analytical techniques such as atomic absorption spectrometry (AAS) [8-13], inductively coupled plasma-optical emission spectrometry [14-16], chromatography [17-19], atomic fluorescence spectrometry [20-22] and X-ray fluorescence spectrometry [23-25] have been used for determination of heavy metals in different samples. However, direct determination of trace metallic ions in environmental and food samples is sometimes difficult due to various factors, particularly their low concentration and matrix effect. Therefore, prior to analysis in complex matrices, a sample preparation step is required for clean-up and/or preconcentration, and to eliminate or minimize matrix effect leading to low detection limits and the improved sensitivity of detection techniques towards analytes. Several sample preparation methods such as liquid-liquid extraction (LLE) [26], coprecipitation [27], cloud point extraction (CPE) [28,29], solid phase extraction [30,31], solid phase microextraction [32], ion-exchange [33], and dispersive solid phase extraction [34,35] have been developed for pretreatment of samples containing heavy metals. Among these developed preconcentration and separation methods, LLE is tedious, multistage operation, and time-consuming. Also, it requires too much of toxic and expensive organic solvents causing health hazards and resulting in the production of hazardous waste. Compared with LLE, SPE has some advantages such as higher enrichment factor (EF), simpler operation, and ease of automation, but relatively high volumes of conditioning, washing, and elution solvents are used. CPE is comparatively cheap, simple, and uses less toxic chemicals. Recently, much attention is being paid to the development of miniaturized, more efficient and environmentally friendly extraction techniques which could greatly reduce the organic solvent consumption. Therefore, liquid-phase microextraction (LPME) methods [36,37] such as single-

drop microextraction (SDME) [38] and hollow fiber liquid-phase microextraction (HF-LPME) [39] were developed as solvent minimized sample pretreatment procedures. Dispersive liquid-liquid microextraction (DLLME) based on a ternary component solvent system, as a fast, simple, and efficient sample preparation method was introduced by Assadi and co-workers [40]. In this microextraction method, very large surface area between an aqueous sample and the fine droplets of an extraction solvent is achievable, and fast extraction kinetic results in the rapid establishment of equilibrium. It was used for the preconcentration and extraction of heavy metals [41-43]. In conventional DLLME, usually 1-2 ml of a disperser solvent is used for dispersing an extraction solvent into an aqueous sample solution. Therefore, the presence of the disperser solvent makes the sample relatively nonpolar leading to high solubility of the lipophilic target analytes in the aqueous sample solution, and results in relatively low extraction efficiency. To solve this problem, some alternatives such as vortex-assisted DLLME [44,45], up-and-down shaker-assisted DLLME [46], air-assisted liquid-liquid microextraction [47-49] and effervescence-assisted-DLLME (EA-DLLME) [50-57] have been proposed.

The aim of this study was to develop a simple and rapid microextraction method that can extract the selected heavy metals based on green EA-DLLME. In the proposed method, phthalic acid is used as complexing agent as well as co-disperser by generating CO_2 from bicarbonate ions. The resulted CO_2 is used in dispersion of the extraction solvent as tiny droplets into the aqueous sample solution and subsequent extraction of the phthalate-cation complexes. Then, the extraction solvent containing the analytes is collected at the bottom of a test tube. Therefore, the proposed EA-DLLME method was performed without using dispersion solvent and additional chelating agent. To investigate the efficiency of the method, the collected organic phase is removed and injected into GFAAS.

EXPERIMENTAL

Apparatus

The measurements were performed with a Shimadzu 6300 absorption spectrometer (Kyoto, Japan) equipped with a heated graphite tube atomizer. The instrument settings and

the optimized furnace program for determination of each analyte are listed in Table 1. An ASC 6100 autosampler (Shimadzu, Kyoto, Japan) was used to deliver standard solutions and samples from the cup to the graphite tube. pH measurements were performed with a Metrohm pH meter model 654 (Herisau, Switzerland). A Hettich centrifuge, model ROTOFIX 32A (Kirchlengern, Germany) was used for accelerating phase separation. Heating of the solution to investigate the effect of temperature was performed by a laboratory heating plate from Gerhardt (Konigswinter, Germany).

Material and Methods

A mixture stock solution of Co(II) and Ni(II) (10 mg l^{-1} of each) was prepared from analytical reagent grade $\text{Co}(\text{NO}_3)_2 \cdot 6\text{H}_2\text{O}$ and $\text{Ni}(\text{NO}_3)_2 \cdot 7\text{H}_2\text{O}$ (Merck, Darmstadt, Germany) by dissolving an appropriate amount of each salt in deionized water (Ghazi Company, Tabriz, Iran). A working standard solution (100 ng l^{-1} of each) was prepared daily by diluting the stock solution with deionized water. Also, a mixture standard solution with a concentration of 0.1 mg l^{-1} of each analyte was prepared and injected into GFAAS each day (three times) for quality control of detection system and the obtained signals were used to calculate enrichment factors (EFs) and extraction recoveries (ERs) of the analytes. Phthalic acid as a complexing agent and co-disperser agent was obtained from Sigma-Aldrich (Seelze, Germany). Sodium hydroxide, sodium bicarbonate, sodium chloride, and the tested extraction solvents were taken from Merck.

Real Samples

River water was from Mehranrood River (Tabriz, Iran). Surface water and well water samples were collected from suburb of Tabriz (East Azarbaijan province, Iran). Peach and orange juice samples were supplied from local supermarkets (in Tabriz, Iran). All samples were centrifuged and the upper phase directly subjected to the extraction method without any pretreatment.

Procedure

Initially 70 mg sodium bicarbonate was placed in a 10-ml dried glass tube with a conical bottom, and $1,1,2,2\text{-tetrachloroethane}$ ($1,1,2,2\text{-TCE}$) as the extraction solvent

($37 \mu\text{l}$) was added to it. In another test tube, 130 mg phthalic acid (as complexing agent) and 400 mg NaCl were added to 5.0 ml of standard solution or real sample, and it was heated at $80 \text{ }^\circ\text{C}$ for 5 min to dissolve the phthalic acid and form complex with the cations. After decreasing the temperature to $45 \text{ }^\circ\text{C}$, the contents of the second tube were transferred into the first tube by a syringe. In the presence of aqueous solution, a reaction between excess amount of phthalic acid and sodium bicarbonate was instantly occurred and the resulted CO_2 led to dispersion of the extraction solvent as tiny droplets into the aqueous solutions and a cloudy solution was formed. After 2 min , the tube was centrifuged at $2191 \times g$ for 7 min , and the fine droplets of organic phase containing the extracted cation-phthalate complexes were sedimented at the bottom of the tube. Its volume ($25 \pm 1 \mu\text{l}$) was measured using a $50\text{-}\mu\text{l}$ microsyringe (zero dead volume, Hamilton, Switzerland). Overall time of the procedure was less than 20 min . In order to investigate the extracted amount of the analytes, two $10\text{-}\mu\text{l}$ aliquots of the settled phase were removed and separately injected into GFAAS. The extraction procedure and the setup used in this method are shown in Fig. 1

RESULTS AND DISCUSSION

In order to investigate the experimental conditions of the proposed method affecting the extraction efficiency, different parameters should be optimized. It is noted that optimization of the procedure was carried out using 5 ml deionized water spiked with 100 ng l^{-1} of each heavy metal.

Effect of Phthalic Acid and Sodium Bicarbonate Amounts

In this study, phthalic acid has double roles: a) as a complexing agent (Fig. 2) and b) as a co-disperser. The sodium bicarbonate and phthalic acid amounts can affect the intensity of the gas formation, and, consequently, the extraction efficiency. Therefore, to study the effect of this parameter, the ERs of the analytes were investigated in the presence of different weights of phthalic acid and sodium bicarbonate ($90:110$, $110:90$, $120:80$, $130:70$, $140:60$, $150:50$, and $160:400$, $\text{mg} : \text{mg}$, respectively). In all tests, the total weight was selected 200 mg . The results in Fig. 3 show that the ratio of $130:70$ (phthalic acid and sodium

Table 1. Instrument Settings and Furnace Programs for Analysis of Co and Ni by GFAAS

Conditions	Co	Ni
Wavelength (nm)	240.7	232.0
Lamp current (mA)	20	25
Ar flow (ml min ⁻¹)	250	250
Injection volume (μl)	10	10
Heating program temperature °C [ramp time (s), hold time (s)]		
Drying 1	110 (20, 0)	110 (20, 0)
Drying 2	250 (10, 0)	250 (10, 0)
Pyrolysis 1	1000 (10, 0)	1000 (10, 0)
Pyrolysis 2	1300 (0,13)	1300 (0,13)
Atomization	2300 (0, 2)	2150 (0, 2)
Cleaning	2400 (0, 2)	2250 (0, 2)

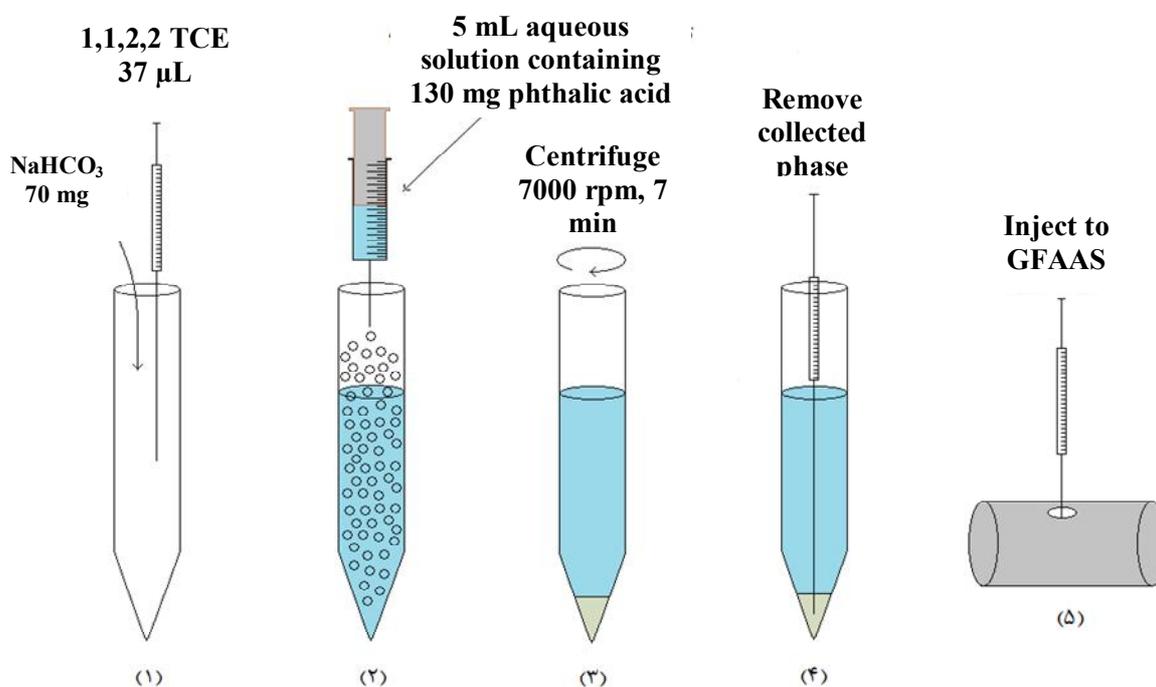


Fig. 1. Extraction and preconcentration procedure.

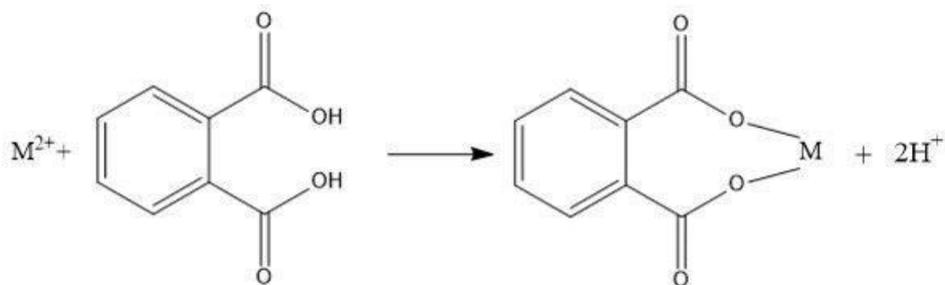


Fig. 2. Reaction of the selected heavy metals with phthalic acid.

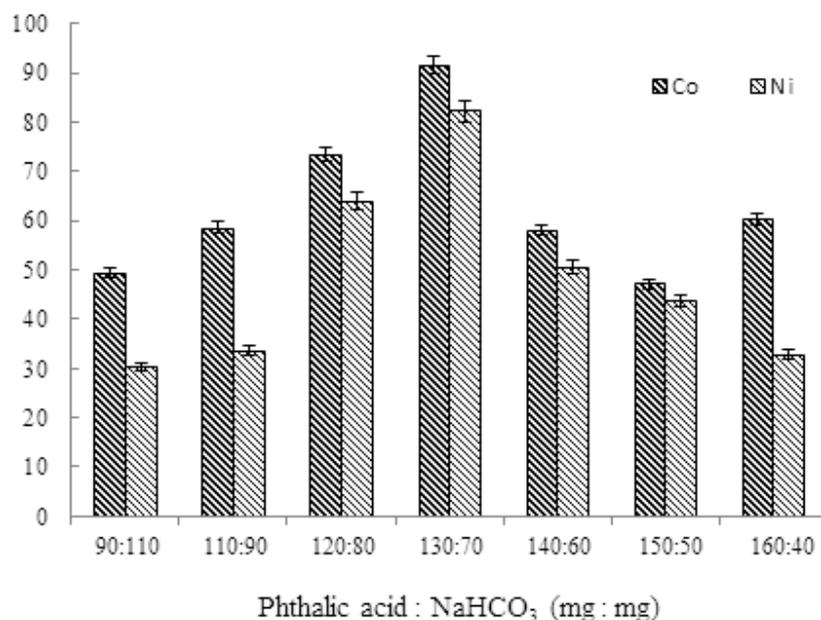


Fig. 3. Effect of phthalic acid and sodium bicarbonate amount on the ER of the analytes. Extraction conditions: sample, 5 ml deionized water containing 100 ng l⁻¹ of Co²⁺ and Ni²⁺ (each cation); extraction solvent (volume), 1,2-DBE (47 μl); temperature, 20 °C; centrifuge rate, 1118 × g; and centrifuge time, 5 min. The error bars represent standard deviations (n = 3).

bicarbonate, respectively) is the most effective composition and gives the highest extraction efficiency. The total weight of sodium bicarbonate and phthalic acid is another important factor affecting the complexation, production the CO₂ gas, and dispersion of the extraction solvent. By taking into the obtained optimized ratio of phthalic acid:sodium bicarbonate (13:7) in the previous step, the effect of the total weight of these compounds was investigated in the range of 100-250 mg. According to the results, up to 200 mg, ERs were increased and thereafter gradually decreased.

Decreasing extraction efficiency at high amounts is probably due to high intensity of the CO₂ formation and quickly removes the gas from the solution. Therefore, 200 mg was selected as the optimum total weight. To carry out the following experiments, 130 mg phthalic acid was dissolved in the solution and 70 mg sodium bicarbonate was placed in the another test.

Selection of Extraction Solvent

Extraction solvent nature is one of the most important

factors in all microextraction methods. Selection of the extraction solvent in the proposed method was performed by considering requirements including formation of microdroplets in the presence of the gas disperser, high extraction capacity toward the analytes, low solubility in water, no reaction with phthalic acid and sodium bicarbonate, and differ density from water. Based on these requirements, different organic solvents including toluene (85 μ l), n-pentadecane (35 μ l), mesitylene (44 μ l), 1,2-dibromoethane (47 μ l), and 1,1,2,2-tetrachloroethane (37 μ l) were tested. It is noted that in all cases a same volume (25 ± 1 μ l) of the collected organic phase was obtained. Figure 4 shows the effect of extraction solvent nature on the ERs of the analytes. Based on these results, 1,1,2,2-TCE and 1,2-DBE are the most effective extraction solvents giving the highest extraction efficiency for the target analytes among the five solvents investigated. Due to low consumption of 1,1,2,2-TCE compared to 1,2-DBE (37 vs. 47 μ l) and its low toxicity, it was selected as the extraction solvent for the further experiments.

Effect of Extraction Solvent Volume

The volume of extraction solvent is another important factor affecting the analytical signals. Increasing 1,1,2,2-TCE volume would increase the extracted amounts of Co(II) and Ni(II), whereas their concentrations in the collected phase will be diluted. Therefore, the effect of volume of 1,1,2,2-TCE was evaluated with several experiments using 30, 37, 50 and 60 μ l 1,1,2,2-TCE while the other parameters were kept constant. These experiments showed that in the volume of 30 μ l of 1,1,2,2-TCE, the volume of the collected phase was 18 μ l, and it was less than 20 μ l, by which the analysis of two analytes was impossible (10 μ l was required for analysis of each cation). Also, when >37 μ l 1,1,2,2-TCE was employed, the analytical signals decreased. This is because of increasing the collected organic phase volume and dilution of the extracted analytes. Hence, a volume of 37 μ l was selected as the optimal value for 1,1,2,2-TCE to carry out the subsequent steps.

Effect of pH

Extraction of the studied cations by the proposed EA-

DLLME method involves prior formation of complexes (Co/Ni-phthalate) with sufficient hydrophobicity which are able to be extracted into the small dispersed volumes of 1,1,2,2-TCE. Considering the phthalic acid as a dicarboxylic acid, the formation of these complexes should be pH-dependent. To study this effect, the influence of pH on the ERs of the analytes was investigated in the pH range of 2-12. The results in Fig. 5 indicate that the maximum extraction of the analytes is obtained at pH = 6-7. At low pHs, decreasing the ERs of the analytes may be attributed to the interaction of phthalate ions with hydronium ions rather than the analytes (phthalic acid $pK_{a1} = 2.89$, $pK_{a2} = 5.51$). The analytical signals also decrease in the alkaline pHs, probably owing to hydrolysis of the studied cations at those pHs. The pH of all samples was in this range, and therefore, there was no need for further pH adjustment in this study.

Effect of Temperature

Temperature is another effective parameter on the performance of the proposed method. It could affect the complexation ability of phthalic acid as well as its reaction with sodium bicarbonate and the amount of produced CO₂ bubbles affecting the dispersion of 1,1,2,2-TCE. Mass transfer improvement which is achievable at high temperatures, is an important phenomenon and can play a key role in the extraction method. Therefore, the effect of temperature on the ERs of Co(II) and Ni(II) was evaluated in the range of 20-90 °C. As seen in Fig. 6, the ERs increase with increasing the temperature from 20 to 45 °C, and then remain approximately constant up to 60 °C. Decreasing the ERs in the temperatures >60 °C, can be due to faster escape of the produced CO₂ gas from the solution. Hence, calibration graphs and the other analytical characteristics of the proposed method were investigated at 45 °C.

Effect of Ionic Strength

The salting-out effect has been commonly used for the enhancement of extraction efficiency. Generally, salt addition can decrease the solubility of analytes in the aqueous phase (and can also reduce the solubility of organic solvents in water) while enhancing their partitioning into the organic phase. To evaluate this effect, different amounts of sodium chloride (0-15%, w/v) were examined. The results

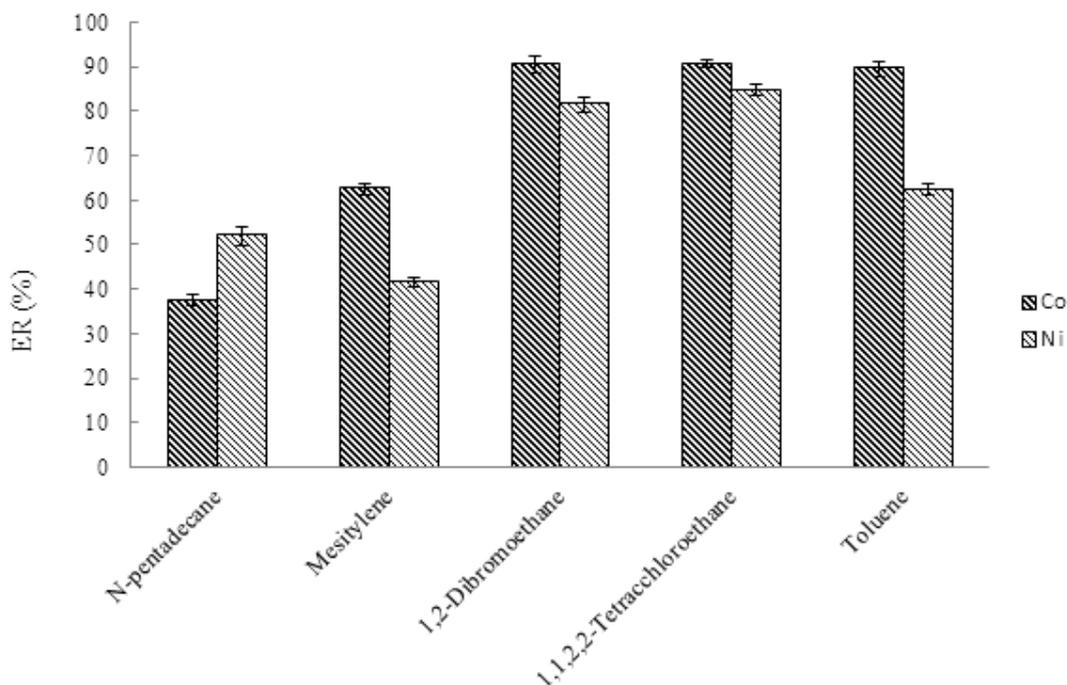


Fig. 4. Effect of extraction solvent type on the extraction efficiency of the method. Extraction conditions: sample, 5 ml deionized water containing 100 ng l^{-1} of Co^{2+} and Ni^{2+} (each cation); phthalic acid; 130 mg, sodium bicarbonate; 70 mg, temperature, $20 \text{ }^{\circ}\text{C}$; centrifuge rate, $1118 \times \text{g}$; centrifuge time, 5 min. The error bars represent standard deviations ($n = 3$).

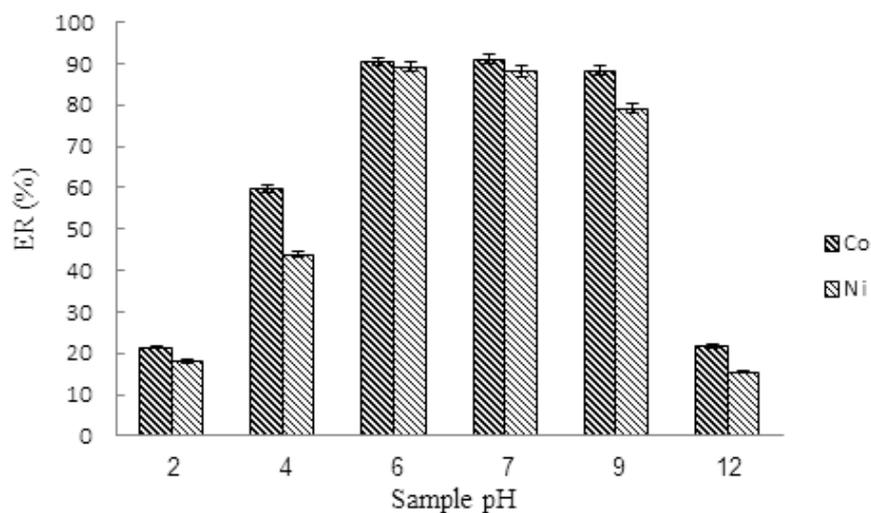


Fig. 5. Study of sample pH on the ERs of the analytes. Extraction conditions: sample, 5 ml deionized water containing 100 ng l^{-1} of Co^{2+} and Ni^{2+} (each cation); phthalic acid; 130 mg, sodium bicarbonate; 70 mg, temperature, $20 \text{ }^{\circ}\text{C}$; extraction solvent (volume), 1,1,2,2-TCE ($37 \text{ } \mu\text{l}$); $20 \text{ }^{\circ}\text{C}$; centrifuge rate, $1118 \times \text{g}$; centrifuge time, 5 min. The error bars represent standard deviations ($n = 3$).

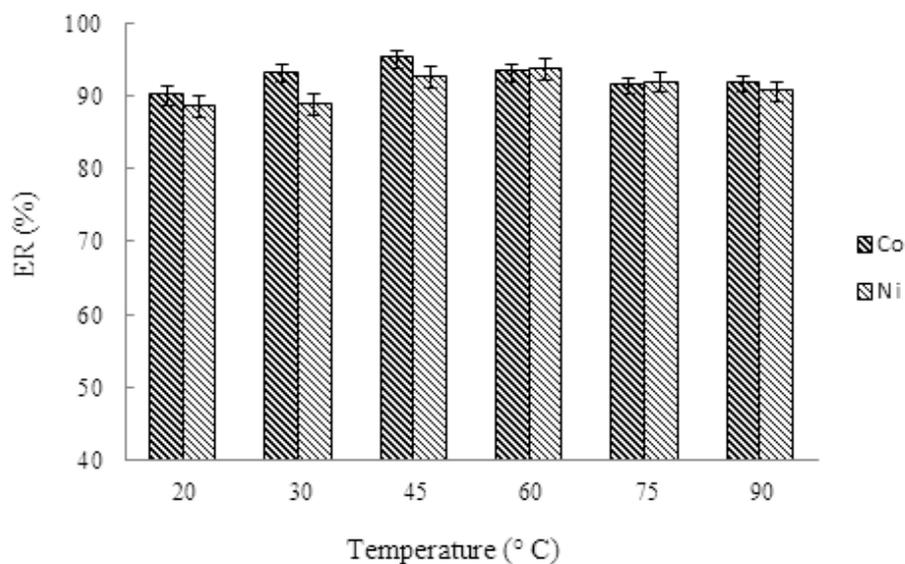


Fig. 6. Effect of temperature on the ERs of the cations. sample, 5 ml deionized water containing 100 ng l⁻¹ of Co²⁺ and Ni²⁺ (each cation); phthalic acid; 130 mg, sodium bicarbonate; 70 mg, extraction solvent (volume), 1,1,2,2-TCE (37 μl); temperature, 20 °C; centrifuge rate, 1118 × g; centrifuge time, 5 min The error bars represent standard deviations (n = 3).

Table 2. Tolerance Limit of Interferent/Analyte Ratios of Coexisting Ions in Determination of Co and Ni (100 ng l⁻¹) by Using the Proposed Method

Species	Tolerance limit of interferent : analyte ratio	
	Co ²⁺	Ni ²⁺
K ⁺	2000	2300
Fe ³⁺	1000	1000
Mg ²⁺	1500	1500
Ca ²⁺	750	2100
Hg ²⁺	500	250
Zn ²⁺	1000	1000
Pb ²⁺	600	200
<u>As³⁺</u>	<u>900</u>	<u>900</u>
<u>Cr⁶⁺</u>	<u>750</u>	<u>1000</u>
Cd ²⁺	250	200
<u>Cl⁻</u>	<u>2000</u>	<u>2000</u>
<u>NO³⁻</u>	<u>1500</u>	<u>1500</u>
CO ₃ ²⁻	1125	3150
SO ₄ ²⁻	1470	1470

showed that with increasing the concentration of NaCl up to 8%, ERs were increased slightly and then remain constant. Regarding the increase in extraction efficiency with the increase in NaCl concentration, the salting-out effect is thought to be responsible. Addition of salt increases the ionic strength of the samples and makes the cation-phthalate complexes less soluble and forces them to migrate into the organic phase. Therefore, 8% (w/v) NaCl concentration was chosen as the optimum salt concentration for the further experiments.

Effect of Centrifuging Conditions

The effect of time and speed of centrifuging were examined in the ranges of 1-10 min and 45-2862 × g, respectively. Therefore, two series of experiments were carried out. In one experiment a constant centrifugation time (5 min) was selected while its speed varied in the range 45-2862 × g. Another experiment was performed at optimized centrifugation speed (2191 × g) while the run time varied (1-10 min). From the results obtained, speed and time of centrifugations were selected as 2191 × g and 7 min, respectively.

Effect of Coexisting Ions

The effect of common coexisting ions in natural water samples on the ER of the analytes was studied. In these experiments, 5 ml of solution containing 100 ng l⁻¹ of the analytes and various amounts of interfering ions was treated according to the developed procedure. A given species was considered to interfere if it resulted in a ± 5% variation in the absorbance signals. The tolerable concentration ratios of the coexisting ions to the analytes are shown in Table 2. The results show that the developed method is selective and can be used without significant interferences from the mentioned coexisting cations and anions.

Analytical Figures of Merit

Quantitative characteristics of the proposed method were obtained in the seven standard solutions under the optimized conditions, and each concentration was injected for two times. Some quantitative parameters including linear range (LR), limit of detection (LOD), limit of quantification (LOQ), coefficient of determination, precision expressed as relative standard deviation (RSD),

EF and ER were evaluated in order to determine efficiency of the method in analysis of Co(II) and Ni(II) in aqueous samples. It is observed that RSD values were equal or less than 3% for intra- and inter-day precisions indicating that an acceptable repeatability is achievable for the developed method. The LODs calculated as 3S_B/m (S_B and m are the standard deviation of the blank and the slope of the calibration graph, respectively) were obtained 9.2 and 15.0 ng l⁻¹ for Co(II) and Ni(II), respectively. EF is defined as the ratio between the analyte concentration in the sedimented phase (C_{sed}) and the initial concentration of the analyte (C₀) in the sample:

$$EF = C_{sed}/C_0 \quad (1)$$

C_{sed} is obtained from a calibration graph. ER is defined as the percentage of the total analyte amount (n₀) which is extracted into the sedimented phase (n_{sed}),

$$ER = \frac{n_{sed}}{n_0} \times 100 = \frac{C_{sed} \times V_{sed}}{C_0 \times V_{aq}} \times 100 = EF \times \frac{V_{sed}}{V_{aq}} \times 100 \quad (2)$$

where V_{col} and V_{aq} are the volumes of the collected phase and aqueous solution, respectively. Enhancement factor (EnF) is also defined as the ratio between the slope of the calibration graph obtained after performing the developed procedure and the slope of the calibration graph obtained by direct injection of the heavy metal solution without extraction. The results are summarized in Table 3.

The accuracy of the developed method was assessed with the measurement of the analytes in NRCC-SLRS-4 as the certified reference material. For analysis of Ni(II) in the certified reference material, it was diluted 5 times with deionized water and then subjected to the presented procedure. The certified and observed values are shown in Table 4. It was found that the results analyzed are in good agreement with the certified values.

Comparison of the Developed Method with other Approaches

Comparative merit of the proposed method and those of the other published methods including the values of LR, LOD, RSD and EF for analysis of the studied cations in different matrices are summarized in Table 5. As seen, the

Table 3. Quantitative Characteristic of the Proposed Method for the Analysis of Co(II) and Ni(II)

Analyte	Regression equation (proposed method)	Regression equation (direct determination)	LR (ng l ⁻¹) ^a	R ² ^b	LOD (ng l ⁻¹) ^c	LOQ (ng l ⁻¹) ^d	RSD% (n = 6) ^e						ER± SD ^f	EF± SD ^g	EnF ± SD ^h
							Intra-day (ng l ⁻¹)			Inter-day (ng l ⁻¹)					
							100	250	500	100	250	500			
Co ²⁺	$Y = 4.12 \times 10^{-4}X - 1.3 \times 10^{-5}$ ⁱ	$Y = 1.48 \times 10^{-6}X - 1.7 \times 10^{-5}$	25.0-1000.0	0.992	9.2	20.0	1.0	1.1	1.0	1.8	1.7	1.5	98.9 ± 1.1	197.8 ± 2.2	278.4 ± 6.2
Ni ²⁺	$Y = 1.6 \times 10^{-4}X + 8 \times 10^{-5}$	$Y = 6.2 \times 10^{-7}X - 8.0 \times 10^{-4}$	50.0-1000.0	0.995	15.0	40.0	1.3	1.3	1.2	3.0	2.8	2.5	98.2 ± 1.3	196.4 ± 2.6	258.1 ± 7.9

^aLinear range. ^bCoefficient of determination. ^cLimit of detection. ^dLimit of quantification. ^eRelative standard deviation (C = 100 ng l⁻¹ of each analyte, n = 6). ^fExtraction recovery ± standard deviation (n = 3). ^gEnrichment factor ± standard deviation (n = 3). ^hEnhancement factor ± standard deviation (n = 3). ⁱX = Concentration (ng l⁻¹) and Y = absorbance.

Table 4. Analysis of the Certified Reference Material (NRCC-SLRS-4) for the Determination of Co^{2+} and Ni^{2+} with the Proposed EA-DLLME Method

Cation	Certified value (ng l^{-1}) \pm S.D. (n = 3) ^a	Found (ng l^{-1}) \pm S.D. (n = 3)	<i>t</i> test ^b
Co^{2+}	33 \pm 8	37 \pm 1	2.079
Ni^{2+}	670 \pm 80	641 \pm 2	1.635

^aStandard deviation. ^b $t_{0.05,2} = 2.920$.

Table 5. Comparison of the Proposed Method with the other Methods Used in Preconcentration and Determination of Co^{2+} and Ni^{2+}

Analyte	Sample preparation method/ Detection system	LR (ng l^{-1}) ^a	LOD (ng l^{-1}) ^b	RSD (%) ^c	Sample volume (ml)	EF ^d	AR (%) ^e	RR (%) ^f	Ref.
Co^{2+} Ni^{2+}	DLLME/GFAAS ^g	- -	21 33	7.5 8.2	5.0	101 200	=	<u>90-109</u>	[58]
Co^{2+} Ni^{2+}	CHE-HLLE/GFAAS ^h	80-1000 40-700	31.8 15.3	5.1 5.2	5.0	- -	=	<u>88-98</u>	[59]
Co^{2+} Ni^{2+}	Emulsion/GFAAS	- -	6 10	- -	100.0	100 100	-	-	[60]
Co^{2+}	UA- SFODME/GFAAS ⁱ	-	20	2.8	20.0	-	=	<u>94-111</u>	[61]
Ni^{2+}	HF-LPME/ETAAS ^j	-	30	4-5	3.0	60	=	<u>95-105</u>	[62]
Co^{2+}	UARS-CPE/W-Coil- ET-AAS ^k	-	300	4.1	10	52	=	<u>93.8-106</u>	[63]
Ni^{2+}	DLLME/ETAAS	-	40	2.1	5.0	138	=	=	[64]
Ni^{2+}	DLLME/GFAAS	-	18	6.5	-	67	=	=	[65]

104

150

Table 5. Continued

Co ²⁺	DLLME/FO-LADS ^l	1-70 µg l ⁻¹	200	<4	10.0	165	=	97-105	[66]
Co ²⁺	MSPE/FL-ICP-OES ^m	2500-100000	800	3.9	60	93	=	90-110	[67]
Ni ²⁺		2500-100000	800	4.9		86	=		
Co ²⁺	SS-LPME/FAAS ⁿ	2500-250000	720	2.9		101.1	=	98.0-99.6	[68]
Ni ²⁺		2500-250000	760	3.7		102.5	=		
Ni ²⁺	RSCPE ^o /FAAS	2000-200000	600	2.3	25	45	=	99.0-103.8	[69]
Co ²⁺	MOFs/ICP-AES ^p	2500-100000	1000	3.9	185	83	22.43	88-104	[70]
Ni ²⁺		2500-100000	900	6.4		92	24.86		
Co ²⁺	PA-	25.0-1000.0	9.2	1.0	5.0	197.8	98.9	89.8-98.4	Present work
Ni ²⁺	LLME/GFAAS ^q	50.0-1000.0	15.0	1.3		196.4	98.2		

^a Linear range. ^b Limit of detection. ^c Relative standard deviation. ^d Enrichment factor. ^e Absolute recovery. ^f Relative recovery. ^g Dispersive liquid liquid microextraction-graphite furnace atomic absorption spectrometry. ^h Cyclohexylamine-homogeneous liquid liquid microextraction-graphite furnace atomic absorption spectrometry. ⁱ Ultrasound assisted microextraction based on solidification of floating organic drop-graphite furnace atomic absorption spectrometry. ^j Hollow fiber liquid-phase microextraction-electrothermal atomic absorption spectrometry. ^k Ultrasound-assisted rapidly synergistic cloud point extraction-tungsten coil electrothermal atomic absorption spectrometer. ^l Dispersive liquid liquid microextraction-fiber optic-linear array detection spectrophotometry. ^m Magnetic solid phase extraction-flow injection inductively coupled plasma optical emission spectrometry. ⁿ Switchable solvent based on liquid phase microextraction-flame atomic absorption spectrometry. ^o Rapidly synergistic cloud point extraction. ^p metal-organic frameworks-inductively coupled plasma atomic emission spectrometry. ^q Phthalic acid based liquid liquid microextraction-graphite furnace atomic absorption spectrometry.

136

137 LODs and RSDs of the proposed method are superior to
138 those of most reported methods except “Emulsion/GFAAS”
139 method. Also, the EFs of the proposed method are better
140 than those reported for the other methods. These results
141 reveal that the presented method is sensitive, simple, rapid,
142 and repeatable and can be used for preconcentration and
143 determination of ultra-trace of Co(II) and Ni(II) from
144 aqueous samples.

145

146 Real Samples Analysis

147 To evaluate applicability of the developed method, it
148 was used for the analysis of several real samples. All
149 samples were extracted and analyzed with the optimized

182

183 procedure. Relative recoveries (RRs) for Co(II) and Ni(II)
184 in real samples in comparison with the results obtained for
185 deionized water spiked at the same concentrations (30, 50,
186 and 100 ng l⁻¹ of Co²⁺ and 60, 100, and 150 ng l⁻¹ of Ni²⁺)
187 are summarized in Table 6.

188

$$189 \text{ RR (\%)} = (\text{Found concentration for a specified} \\ 190 \text{ concentration spiked in real sample}) / (\text{Found} \\ 191 \text{ concentration for the same spiked} \\ 192 \text{ concentration in deioni} \times 100) \quad (3)$$

193

194 The obtained recoveries are between 89.8 and 98.4%
195 indicating that matrices of the real samples have a little

Table 6. Results of Assays to Check the Samples Matrices Effect for the Selected Cations and Concentrations of the Detected Analytes

Analyte	Spiked concentration (ng l ⁻¹)	River water		Surface water		Well water		Peach juice		Orange juice	
		Found concentration	Relative recovery ± S.D. (n = 3)	Found concentration	Relative recovery ± S.D. (n = 3)	Found concentration	Relative recovery ± S.D. (n = 3)	Found concentration	Relative recovery ± S.D. (n = 3)	Found concentration	Relative recovery ± S.D. (n = 3)
Co ²⁺	0	609.2 ± 9.0	-	273.4 ± 4.9	-	N.D. ^a	-	N.D.	-	N.D.	-
	<u>30.0</u>	<u>638.2 ± 7.4</u>	<u>96.8 ±</u>	<u>94.3 ±</u>	<u>27.4 ± 0.4</u>	<u>91.4 ±</u>	<u>27.7 ± 0.4</u>	<u>92.2 ±</u>	<u>27.1 ± 0.4</u>	<u>90.3 ±</u>	
			<u>1.2</u>	<u>1.2</u>		<u>1.3</u>	<u>1.4</u>	<u>1.4</u>			
	50.0	656.8 ± 9.0	95.2 ±	91.8 ±	47.9 ± 0.8	95.9 ±	46.5 ± 0.7	92.9 ±	44.9 ± 0.7	89.8 ±	
			1.3	1.5		1.6	1.4	1.6			
100.0	707.6 ± 7.9	98.4 ±	93.3 ±	97.8 ± 1.5	97.8 ±	94.2 ± 1.6	94.2 ±	92.4 ± 1.6	92.4 ±		
		1.1	1.3		1.5	1.6	1.6				
Ni ²⁺	0	55.3 ± 0.9	-	94.7 ± 1.7	-	141.1 ± 3	-	N.D.	-	N.D.	-
	<u>60.0</u>	<u>112.0 ± 1.6</u>	<u>94.5 ±</u>	<u>95.5 ±</u>	<u>197.9 ± 3.1</u>	<u>94.7 ±</u>	<u>55.8 ± 0.7</u>	<u>93.0 ±</u>	<u>56.3 ± 0.8</u>	<u>93.8 ±</u>	
			<u>1.4</u>	<u>1.5</u>		<u>1.6</u>	<u>1.2</u>	<u>1.3</u>			
	100.0	152.6 ± 2.7	97.3 ±	97.8 ±	235.6 ± 3.5	94.5 ±	96.2 ± 1.5	96.2 ±	94.9 ± 1.4	94.9 ±	
			1.7	1.6		1.4	1.6	1.4			
<u>150.0</u>	<u>201.6 ± 3.4</u>	<u>97.5 ±</u>	<u>97.2 ±</u>	<u>283.2 ± 4.1</u>	<u>94.7 ±</u>	<u>144.2 ± 2.1</u>	<u>96.1 ±</u>	<u>142.5 ± 2.0</u>	<u>95.0 ±</u>		
		<u>1.7</u>	<u>1.6</u>		<u>1.4</u>	<u>1.4</u>	<u>1.3</u>				

effect on the developed method. Also the obtained concentrations for the standard cations in the sample are listed in Table 6.

CONCLUSIONS

In this study, an effervescence-assisted dispersive liquid-liquid microextraction was proposed for the extraction and preconcentration of the selected heavy metal cations in water and fruit juice samples before their analysis by GFAAS. For the first time, phthalic acid was simultaneously used for complexation of analytes and as co-disperser in DLLME. Also, the CO₂ gas resulting from the reaction of phthalic acid and bicarbonate ions was used as a disperser. Therefore, DLLME was performed without addition of toxic complexing agent and disperser solvent. In the proposed method, much low of an organic solvent (μ -level) was used as the extraction solvent in the absence of disperser solvent for 5 ml of the sample. The method allowed high preconcentration of the analytes owing to the usage of large volumes of samples in a short time. The linearity, ER, EF, and repeatability of the method were validated within the acceptable ranges.

ACKNOWLEDGMENTS

The authors would like to thank the Research Office at the University of Tabriz for financial support.

REFERENCES

- [1] M. Amjadi, A. Samadi, *Colloid. Surface* 434 (2013) 171.
- [2] C.L. Dunlap, S.K. Vincent, B.F. Barker, *J. Am. Dent. Assoc.* 118 (1989) 449.
- [3] A.L. Greppi, D.C. Smith, D.G. Woodside, *Univ. Tor. Dent. J.* 3 (1989) 11.
- [4] WHO 2008, *Guidelines for Drinking Water Quality*, World Health Organization, Geneva.
- [5] V.P. Kudesia, *Water Pollution*, Pregati Prakashan Publications, Meerut, 1990.
- [6] M. Gebrekidan, Z. Samuel, *Mekelle University* 3 (2011) 105.
- [7] V. Okumuş, S. Özdemir, E. Kılınç, A. Dündar, U. Yüksel, Z. Baysal, *Bioremediat. J.* 19 (2015) 47.
- [8] D.V. Babos, A.I. Barros, E.C. Ferreira, J.A.G. Neto, *Spectrochim. Acta B* 130 (2017) 39.
- [9] M. Fayazi, M. Ghanei-Motlagh, M.A. Taher, R. Fayazi, *Anal. Bioanal. Chem. Res.* 3 (2016) 87.
- [10] G. Özzeybek, S. Erarpat, D.S. Chormey, M. Firat, Ç. Büyükpınar, F. Turak, S. Bakırdere, *Microchem. J.* 132 (2017) 406.
- [11] F.A. Lobo, D. Goveia, A.P. Oliveira, L.P.C. Romão, L.F. Fraceto, N.L.D. Filho, A.H. Rosa, *Fuel* 90 (2011) 142.
- [12] H. Abdolmohammad-Zadeh, M. Ahari Salmasi, *Anal. Bioanal. Chem. Res.* 5 (2018) 23.
- [13] I. Calle, F.P. Pereira, I. Lavilla, C. Bendicho, *Anal. Chim. Acta* 936 (2016) 12.
- [14] L. Fu, Sh. Shi, X. Chen, H. Xie, *Microchem. J.* 139 (2018) 236.
- [15] B. Feist, B. Mikula, *Food Chem.* 147 (2014) 302.
- [16] A. Ilander, A. Väisänen, *Anal. Chim. Acta* 602 (2007) 195.
- [17] S.L. Stegall, K.M. Ashraf, J.R. Moye, D.A. Higgins, M.M. Collinson, *J. Chromatogr. A* 1446 (2016) 141.
- [18] M.A. Farajzadeh, A. Yadeghari, *J. Ind. Eng. Chem.* 59 (2018) 377.
- [19] V. Radchenko, J.W. Engle, J. J. Wilson, J.R. Maassen, F.M. Nortier, W.A. Taylor, E.R. Birnbaum, L.A. Hudston, K.D. John, M.E. Fassbender, *J. Chromatogr. A* 1380 (2015) 55.
- [20] J. Zhang, J. Fang, X. Duan, *Spectrochim. Acta B* 122 (2016) 52.
- [21] Z. Lei, L. Chen, K. Hu, Sh. Yang, X. Wen, *Spectrochim. Acta A* 203 (2018) 522.
- [22] Sh. Li, M. Wang, Y. Zhong, Z. Zhang, B. Yang, *Spectrochim. Acta B* 111 (2015) 74.
- [23] R. Hennekam, T. Sweere, Rik Tjallingii, G.J. Lange, G.J. Reichart, *Quatern. Int.* 2018, In Press.
- [24] O.N. Almeida, F.H.M. Luzardo, F.A.C. Amorim, F.G. Velasco, L.N. González, *Spectrochim. Acta B* 150 (2018) 92.
- [25] E. Kamilari, K. Farsalinos, K. Poulas, Ch.G. Kontoyannis, M.G. Orkoula, *Food Chem. Toxicol.* 116 (2018) 233.

- [26] S.Y. Choi, V.T. Nguyen, J. Lee, H. Kanga, B.D. Pandey, *J. Hazard. Mater.* 278 (2014) 258.
- [27] M. Soylak, H. Balgunes, *J. Hazard. Mater.* 155 (2008) 595.
- [28] M.H. Youcef, T. Benabdallah, H. Reffas, *Sep. Purif. Technol.* 149 (2015) 146.
- [29] Ch. Zeng, X. Xu, N. Zhou, Y. Lin, *Spectrochim. Acta A* 94 (2012) 48.
- [30] S. Özdemir, S.A. Mohamedsaid, E. Kılınç, M. Soylak, *Food Chem.* 271 (2019) 232.
- [31] S. Ozdemir, E. Kilinc, E.T. Oner, *Food Chem.* 276 (2019) 174.
- [32] Sh. Su, B. Chen, M. He, B. Hu, *Talanta* 123 (2014) 1.
- [33] L. Dong, L. Hou, Zh. Wang, P. Gu, G. Chen, R. Jiang, *J. Hazard. Mater.* 359 (2018) 76.
- [34] E. Yavuz, Ş. Tokaloğlu, Ş. Patat, *Food Chem.* 263 (2018) 232.
- [35] E. Yavuz, Ş. Tokaloğlu, Ş. Patat, *Talanta* 190 (2018) 397.
- [36] M.A. Farajzadeh, S.M. Sorouraddin, M.R. Afshar Mogaddam, *Microchim. Acta* 181 (2014) 829.
- [37] B. Hu, M. He, B. Chen, L. Xia, *Spectrochim. Acta B* 86 (2013) 14.
- [38] I.H. Šrámková, B. Horstkotte, K. Fikarová, H. Sklenářová, P. Solich, *Talanta* 184 (2018) 162.
- [39] Z. Es'haghi, A. Nezhadali, H.A. Hosseini, S. Mohammadi-Nokhandani, *Arab. J. Chem.* 10 (2017) S3840.
- [40] M. Rezaee, Y. Assadi, M.R. Milani Hosseini, E. Aghaee, F. Ahmadi, S. Berijani, *J. Chromatogr. A* 1116 (2006) 1.
- [41] M. Saraji, H. Ghambari, *J. Chromatogr. A* 1574 (2018) 27.
- [42] J.A. Barreto, R.S. Assis, R. Jorgensen, C. Valfredo, A. Lemos, *Talanta* 193 (2019) 23.
- [43] N. Altunay, E. Yıldırım, R. Gürkan, *Food Chem.* 245 (2018) 586.
- [44] E. Fernández, L. Vidal, A. Costa-García, A. Canals, *Anal. Chim. Acta* 915 (2016) 49.
- [45] R. Akramipour, M.R. Golpayegani, S. Gheini, N. Fattahi, *Talanta* 186 (2018) 17.
- [46] P.Sh. Chen, W.Y. Haung, Sh.D. Huang, *J. Chromatogr. B* 955-956 (2014) 116.
- [47] M.A. Farajzadeh, M.R. Afshar Mogaddam, *Anal. Chim. Acta* 728 (2012) 31.
- [48] S.M. Sorouraddin, M.A. Farajzadeh, S. Nouri, M.R. Afshar Moghaddam, *Anal. Bioanal. Chem. Res.* 4 (2017) 227.
- [49] S.M. Sorouraddin, *J. Iran. Chem. Soc.* 13 (2016) 2219.
- [50] X. Wu, X. Li, M. Yang, H. Zeng, S. Zhang, R. Lu, H. Gao, D. Xu, *J. Chromatogr. A* 1497 (2017) 1.
- [51] X. Liu, Ch. Liu, P. Wang, G. Yao, D. Liu, Zh. Zhou, *Food Chem.* 245 (2018) 653.
- [52] A. Gh. Moghadam, M. Rajabi, M. Hemmati, A. Asghari, *J. Mol. Liq.* 242 (2017) 1176.
- [53] K. Medinskaia, Ch. Vakh, D. Aseeva, V. Andruch, L. Moskvina, A. Bulatov, *Anal. Chim. Acta* 902 (2016) 129.
- [54] Y. Li, J. Hu, W. Liu, L. Jin, P. Zhou, Y. Zhang, B. Zhang, R.A. Dahlgren, X. Wang, Y. Zhou, *Talanta* 195 (2019) 785.
- [55] X. Liu, Ch. Liu, P. Wang, G. Yao, D. Liu, Zh. Zhou, *Food Chem.* 245 (2018) 653.
- [56] A. Shishov, I. Sviridov, I. Timofeeva, N. Chibisova, L. Moskvina, A. Bulatov, *J. Mol. Liq.* 247 (2017) 246.
- [57] G. Lasarte-Aragonés, R. Lucena, S. Cárdenas, M. Valcárcel, *Anal. Chim. Acta* 807 (2014) 61.
- [58] H. Jiang, Y. Qin, B. Hu, *Talanta* 74 (2008) 1160.
- [59] S.M. Sorouraddin, M.A. Farajzadeh, T. Okhravi, *Talanta* 175 (2017) 359.
- [60] H. Matsumiya, T. Kageyama, M. Hiraide, *Anal. Chim. Acta* 507 (2004) 205.
- [61] J. Lan, Z. Zhao, *Chem. Spec. Bioavailab.* 24 (2012) 124.
- [62] J. Abulhassani, J.L. Manzoori, M. Amjadi, *J. Hazard. Mater.* 176 (2010) 481.
- [63] L. Chen, Z. Lei, Sh. Yang, X. Wen, *Microchem. J.* 130 (2017) 452.
- [64] K. Alizadeh, H. Nemati, S. Zohrevand, P. Hashemi, A. Kakanejadifard, M. Shamsipur, M.R. Ganjali, F. Faribod, *Mater. Sci. Eng. C* 33 (2013) 916.
- [65] P. Liang, L. Peng, *Atom. Spectrosc.* 33 (2012) 53.
- [66] N. Shokoufi, F. Shemirani, Y. Assadi, *Anal. Chim. Acta* 597 (2007) 349.
- [67] Y. Yamini, M. Safari, *Microchem. J.* 143 (2018) 503.
- [68] A. Habibiyan, M. Ezoddin, N. Lamei, Kh. Abdi, M.

- Amini, M. Ghazi-khansari, J. Mol. Liq. 242 (2017) 492.
- [69] R. Rahnama, M. Najafi, Environ. Monit. Assess 188 (2016) 150.
- [70] M. Safari, Y. Yamini, M.Y. Masoomi, A. Morsali, A.M. Varnosfaderani, Microchim. Acta 184 (2017) 1555.