A Univariate Optimization Strategy for Pre-concentration of Cobalt(II) in Various Matrixes by a DLLME before Analysis Using FAAS

Zaman Sahb Mehdi[1,2*] and Saher Abdel Reda Ali Alshamkhawy[1]

[1]*Department of Chemistry, College of Science, University of Thi-Qar, Thi-Qar 64001, Iraq*

[2]*Department of Chemistry, College of Science, University of Al-Muthanna, Al-Samawah 58002, Iraq*

*** Corresponding author:***

email: zaman.mehdi@mu.edu.iq

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Abstract: *A procedure based on dispersive liquid-liquid microextraction (DLLME) for cobalt (Co) quantification in an Iraqi environmental matrix by flame atomic absorption spectroscopy (FAAS) was applied in this work. A case-study approach was chosen to obtain further in-depth information on the Co levels and to evaluate the effectiveness of* N-*salicylideneaniline (SAN) as a complexing agent for pre-concentration and extraction of Co. An univariate strategy was utilized to achieve the optimum extraction conditions. The estimated limits of detection (LOD) and quantification (LOQ) under optimum conditions were 1.04 and 3.47 µg* L^{-1}*, respectively. The results achieved by the proposed system were compared with those using the microwave digestion/graphite furnace atomic absorption spectrometer (MWD/GF-AAS) for digest samples and also for some water samples (Direct GF-AAS). The proposed procedure was applied for analyzing eleven environmental samples. The detectable Co levels for water samples ranged from 0.72 to 4.30 µg* L^{-1} *with a relative standard deviation of 3.7–8.8%, while the concentration for solid samples ranged from 0.17–4.51 µg* g^{-1} *(2.4–11.8 RSD %). DLLME/FAAS proposed procedure is effective, simple, and has the benefit of minimizing the organic solvent consumption by a few microliters, which results in little waste.*

Keywords: *bivalve molluscs; cobalt; environmental samples; Schiff base; solvent microextraction*

■ INTRODUCTION

Environmentalists request analytical chemists to investigate the complicated relations between trace metal levels and living organisms by focusing current research on detecting trace metal levels in natural water and vegetables [1-2]. The content of these samples is quite diverse, and the origin of their parent matrix is essential for establishing the strategy and techniques utilized in a study [3]. As a trace element, cobalt (Co) plays an important function for humans, whose daily needs are only a few milligrams. But, if consumed in large quantities, this can be hazardous to human health [4-5]. Natural sources of Co include water, air, soil, animals, rocks, and plants [6]. With the fast expansion in the usage of Co into the metallurgical industry for the production of alloys, rechargeable batteries, catalysts, paints, drugs, and ceramics, increasingly of it is released into the environment and entered the food chain [7-8]. Thus, the total Co content in the food and environmental matrixes must be determined precisely and accurately.

Considering that food and water are prominent sources of matrix effects if Co is directly analyzed, digestion of the sample is typically necessary prior to quantification [9]. Taking into account the range of Co amount often present in food and water [9-10], as well as the inherent dilution produced by digestion of sample [11], high sensitivity instruments [12-13] such as plasma atomization-based ones are frequently required to detect Co in food and water. Nevertheless, flame atomic absorption spectroscopy (FAAS) is the most popular method, and it is employed for the determination of water Co content in soil and food [14]. In addition, FAAS was selected as a reference technique for the validation of Co quantification with UV–vis spectrophotometer [15]. Another atomic absorption spectroscopy method, Flameless atomization, has been utilized for Co quantification with detection limits comparable to plasma techniques, but these instruments are relatively expensive, and their operation and maintenance costs are also substantial [16].

Often, quantifying very low levels of an element requires a separation step with pre-concentration. In addition, the analytical techniques available cannot present the selectivity and sensitivity requirement appropriate for analyzing this form of matrix sample [17-18]. Liquid-liquid extraction (LLE) has been the technique of choice for sample enrichment due to this procedure, among others, has a number of attractive features, such as versatility and simultaneous extractor of several components, as there is the possibility of using a wide variety of organic solvents [19]. However, LLE utilizes large volumes of these solvents, which can be flammable or toxic. Recently, a variety of extraction strategies focused on liquid phase microextraction (LPME) have been developed for the sample's enrichment [20]. A major advantage of these approaches includes a variety of sample preparation techniques aimed at considerably reducing the amount of reagents and solvents [21-22]. Many researchers have highlighted dispersive liquid–liquid microextraction (DLLME) for analytes pre-concentration and extraction for subsequent quantification in food and environmental samples [23-24]. The second advantage of using this method is great enrichment factors, rapidity, and simplicity, which is based on dispersive solvents for absolute mass transfer from the aqueous phase [25]. Considering that DLLME has a great pre-concentration factor, its combination with FAAS is an interesting manner for elements quantification. FAAS was selected for its wide availability, validity, low operating cost, selectivity, and convenience with a pre-concentration method (DLLME), appropriate for many matrixes, and it is also free from most interferences [26-27].

In this work, a procedure of DLLME was developed to pre-concentrate Co(II) to be quantified by FAAS. The DLLME method involves complex formation for increased compatibility with extraction solvent and improved extraction efficiency. Salicylideneaniline (SAN) was used as a ligand for complexing with Co(II). This reagent was applied with LLE techniques for the separation of Co [28]. The analytical procedure optimization was obtained from a univariate strategy. Some main parameters, such as the simultaneous change of extraction and disperser solvent volumes, selectivity, pH, and SAN quantity, were evaluated. The DLLME–FAAS method was applied to Co quantification in food and environmental samples.

■ EXPERIMENTAL SECTION

Materials

Reagents

A standard solution of Co(II) with a concentration of 1,000 μg mL^{-1} is supplied with an analysis certificate, traceable to NIST standards, supplied by Chem-Lab (Zedelgem, Belgium), and it's utilized for freshly dilute and spiking experiments. Sodium sulfate, 99.99% Suparpur (Germany) and potassium acetate, 98.0% (Sigma-Aldrich) for buffer pH range: 4.30–5.50. Other Co solution's pH values were adjusted with phosphate salt pH 6.0–7.0 (Merck), Tris-HCl with pH 7.5–8.0 (Scharlau) and ammoniacal with pH 8.5–9.8 buffer solutions. SAN solution 98%, a Schiff base, was used as a chelating agent and was kindly provided by AK Scientific (USA). The solvents were chloroform ($CHCl_3$, Sigma-Aldrich), 1,2-dichloroethane ($C_2H_4Cl_2$, Merck), ethanol (C_2H_6O, Merck), carbon tetrachloride (CCl_4, Riedel-de Haën), acetone (C_3H_6O, J.T. Baker), methanol (CH_3OH, Sigma-Aldrich), acetonitrile (C_2H_3N, Merck), and acetic acid ($C_2H_4O_2$, HiMedia). HNO_3 (free trace metals) was supplied by Sigma-Aldrich and utilized for solid sample preparation by microwave digestion procedure. Ultra-pure water (Supelco) was used to dilute and prepare aqueous solutions. All other chemicals required for this analysis are reagent-grade or better except otherwise stated.

Collection of samples and pre-treatment

Water, plant, vegetables, and molluscs samples were prepared for application and evaluation of the proposed DLLME–FAAS procedure for analysis of real samples. Six kinds of water samples from Al-Muthanna, Southern Iraq. The samples were filtered immediately (No. 42 Whatman paper) after collection in polypropylene bottles previously soaking for 24 h in 10.0% (v/v) purified nitric acid and, before use, rinsed thoroughly with ultrapure water. Two samples of river water are utilized to provide the raw water for the desalination and treatment water system located in Al-Rumaitha, Al-Muthanna, Iraq. A well water sample was collected from the Al-Muthanna desert, as well as tap and potable water samples. River-water bivalve molluscs as a bio-indicator of pollution were collected from the Euphrates River. The vegetables and medicinal herbal samples (n = 4): lettuce, mint (*Mentha piperita*), black seed (*Nigella sativa*), and dill (*Anethum graveolens*) were collected from the local market in Samawah, Iraq. The microwave digestion (MWD) procedure for sample digestion was used before being washed with ultrapure water, dried in an oven, and pulverized it. Approximately 250 mg of pulverized molluscs, 500 mg of lettuce, 2 g of black seed, and 400 mg of mint and dill each sample were submitted to the MWD system. To validate of results and because the Co(II) amount in some original samples was not detected by the proposed procedure, a solution containing a known concentration of Co was spiked to the samples of diverse water, medicinal herb, and bivalve molluscs. Next, the resultant materials were mineralized by MWD. As well, the sample matrix solution was digested in the same manner, and finally, all samples were diluted with ultrapure water at a convenient volume after the pH was adjusted by ammoniacal solution.

Instrumentation

The instrument employed for spectroscopic measurements of Co solutions is an atomic absorption spectroscopy (AAS) equipped D2-lamp system for background correction (AA-7000 Spectrometer, Shimadzu, Japan) during the quantification of Co. A single element source (high-performance Co-hallow cathode lamp for AA-7000 Spectrometer) was operated at 20 mA. The primary wavelength, 240.7 nm, was selected for this work, with 0.2 nm of slit width. The analytical signal was achieved as absorbance with pre-spray and integration times were 5 and 3 s, respectively. The composition of flame, air-acetylene, was set up at 15 L min^{-1} for support gas and 1.6 L min^{-1} for fuel gas.

The quantification of target elements in digests and also some water matrixes was performed by Shimadzu GFA-7000 graphite furnace-AAS features Deuterium (D2) background correction. The GFA-7000 spectrometer was equipped with a programmable sample dispenser (PSD) autosampler. The PSD delivers measured volumes (20 μL) of the sample for the furnace automatically. The spectrometer optics parameters for Co determination in target digests and in some aqueous samples were selected according to the recommendation of the spectrometer manufacturer (as was mentioned in the first paragraph). The matrix modifier, modifier volume, replicates, and measurement mode were conditioned at 750 mg L^{-1} Pd and 400 mg L^{-1} Mg; 5 μL; 3; and peak height, respectively. The heating program of the furnace was also chosen based on the manufacturer's recommendation for the spectrometer, as shown in Table S1.

A Hettich centrifuge carried out the physical separation of two phases (model EPA 200, Germany) for the separation of the sedimented phase in the DLLME procedure and the undissolved real sample parts. The pHmeter (inoLab pH 7110 xylem analytics, WTW, Germany), connected with a combination pH electrode (SenTix 81), has been utilized to adjust and measure the pH value for real and standard samples. A vortex (MX-F model, MRC, UK) was utilized to homogenize the centrifuge tube content before the injection of the solvent mixture.

Procedure

To proceed with the complexion step, 10 mL of a previously prepared sample or a solution containing μg L^{-1} level of Co adjusted the pH value at 9.0 was added to a 15 mL test tube, conical bottom. This solution also contained 6.33% (m/v) of SAN. The mixture was gently stirred for 5 s with a vortex mixer to disperse the ligand solvent uniformly throughout the aqueous solution and to accelerate the complex formation. Thereafter, to proceed with the micro-extraction step, a solvent solution of 60 μL carbon tetrachloride (CTC) and

1000 µL dispersive solvent (ethanol) was prepared and rapidly added into the test tube by a syringe. During injection, the micro-droplets formed were dispersed throughout the tube, and the resultant product was left for 1.0 min to complete the cloudy solution formation. The Co-SAN complex was then extracted in fine droplets of ethanol. Physical separation was achieved by centrifuging the system at 4,000 rpm for 3.0 min. The lower phase, a clear residue, was measured and collected using a 100 Hamilton syringe, (PerkinElmer, Australia). Finally, to determine the Co absorbance by FAAS, the sedimented organic phase was diluted to 0.5 mL with ethanol.

■ RESULTS AND DISCUSSION

Optimization of Complex Formation

The complex produced must be stable in terms of its pre-concentration/separation from an aqueous medium. To develop an efficient Co-Schiff base complex, the parameters (the pH value of buffer, amount of ligand, and periods of mixing) impacting the efficiency of complex formation were carefully optimized. All experiments were carried out at an extraction time of 1 min and centrifugation at 4,000 rpm for 3 min.

The pH level of the sample's has a considerable impact on the efficiency of the DLLME process, as well as the formation and stability of the complex [29]. Accordingly, equal volumes of buffer solutions with a pH range of 2.7–9.8 were added to aqueous standards containing 160 µg L^{-1} Co(II) solution before proceeding the preconcentrated step into the micro-level volume of the sediment phase by DLLME using SAN. In the first set of analyses, at pH 3.0 and 4.3, no difference greater than the absorbance of the blank was observed. A possible explanation for this might be that at low pH, the decomposition of SAN could occur [30]. The results obtained from the analysis of pH (4.7–9.3) are shown in Fig. 1 and as can be seen, the observed increase in absorbance happened from pH 8.0–9.3. In addition, at a pH of more than 9.5, only trace amounts of Co were detected in this experiment. This discrepancy could be attributed to the precipitation of cobalt hydroxide forms at high pH. It can be seen from the closer inspection of the figure that the absorbance value at pH 9.0 reached its optimum extraction of Co. Hence, in subsequent tests, the pH of the Co(II) solutions was adjusted using an ammoniacal solution with a pH of 9.0.

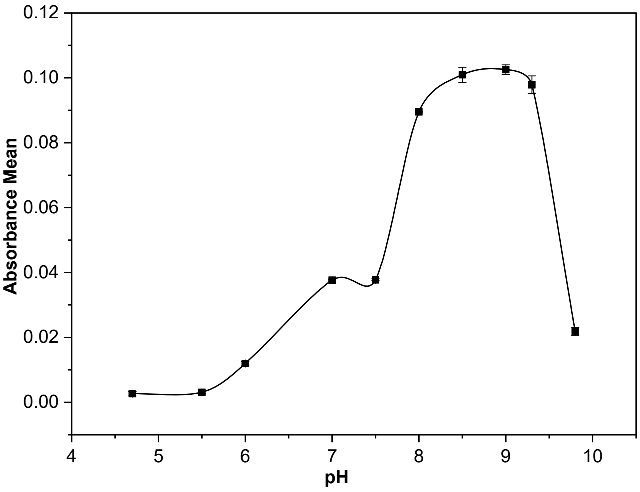

Fig 1. Effect of pH (4.7–9.8) (n = 3) on the absorbance of Co(II) using DLLME–FAAS

SAN was tested as an excellent chelate extractant in LLE for some metals and acted as bidentate ligands in the extraction system [31]. The present procedure utilized SAN as a new complexing agent in Co(II)-DLLME. Therefore, the effect of reagent SAN amount on pre-concentration Co was assessed, changing the concentration (0.050, 0.075, 0.100, 0.200, 0.250, 0.500, 1.000, and 2.000%) of this chelating reagent's solution added into 10.0 mL of the standard solution including, buffered analyte ion at (pH = 9.0), of 1.60 µg Co(II). As shown in Fig. 2, the 0.25% gave the best analytical signal. To volume higher than this percent, as the concentration of SAN was raised further, higher than 0.50%, the absorbance of Co(II) decreased. This discrepancy could be attributed to the excess SAN being extracted. Thus, this reduces the organic solvent's finite ability to extract the Co(II)-SAN complex quantitatively, minimizing microextraction efficiency [32]. Therefore, a 120 µL of 0.25% (m/v) SAN solution prepared with an ethanol solvent was selected as the best value for further studies.

Kind and Volume of Extraction/Disperser Solvents

The use of SAN as a chelating agent produces a stable complex with several metals that can be extracted

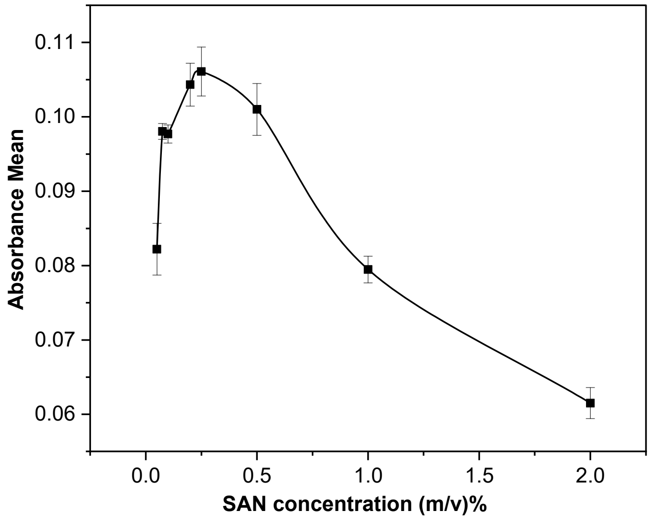

Fig 2. Effect of SAN concentration (0.05–2.0%, m/v) (n = 3) on the absorbance of Co(II) using DLLME–FAAS

by chlorinated solvents [28]. Twelve combinations of disperser (acetone, acetonitrile, ethanol, and methanol) and extraction solvents: 1,2-dichloroethane (DCE), chloroform (CHF), and carbon tetrachloride (CTC) were evaluated.

To estimate the sedimented phase formation, experiments were performed using 1000 µL and 60, 115, and 140 µL of disperser and extraction solvents (CTC, CHF, and DCE), respectively, to obtain a steady sedimented phase. Comparing the results of all experiments, it can be seen that all solvents revealed that centrifugation did lead to the separation of the sedimented phase. The tiny droplets were clearly observed when CTC and CHF were used as extraction solvents. In this experiment, the selection of extraction solvent is based on the maximum recovery of the sample target, peak highest, combined with the lowest standard deviation (RSD%) and blank values. As shown in Fig. 3, CTC and CHF showed the best signals, also the blank values for CTC and DCE solvents were lower (the value of RSD% for CTC experiments, n = 3, has been found to be 1.64). Therefore, CTC solvent was selected for future investigations.

The next section in this investigation was concerned with the study of the amount of extraction solvent, CTC, which was adjusted to achieve maximum DLLME efficiency while minimizing volume. The procedure for the Co(II)–SAN was performed using 35–150 µL CTC. In summary, these results show that the absorbance value is greater when the CTC volume is 60 µL (Fig. 4). Consequently, 60 µL of CTC was used for subsequent tests.

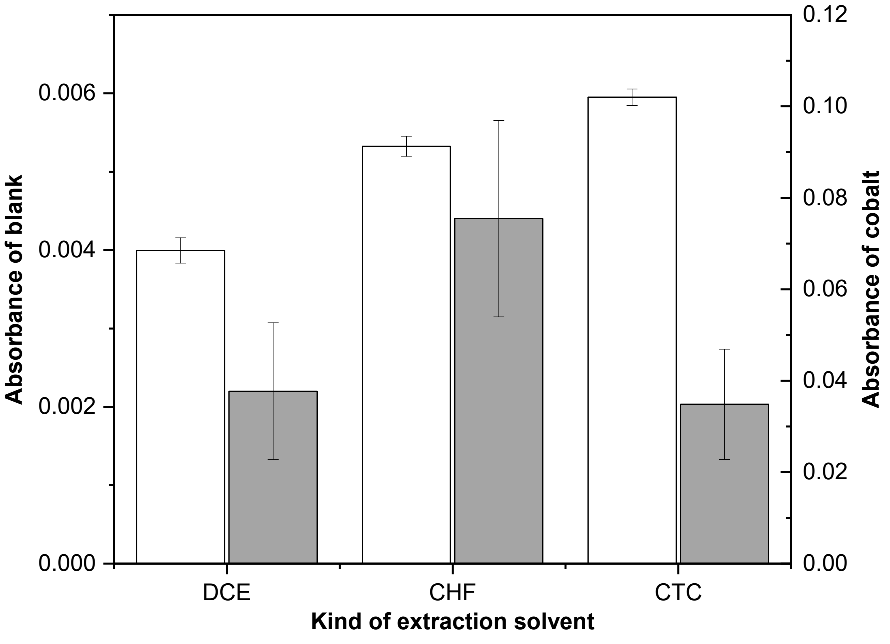

Fig 3. Effect of kind of extraction solvent (DCE, CHF, CTC) (n = 3) on the absorbance of Co(II) using DLLME–FAAS. White column/bar corresponds to the absorbance for Co while the grey bar corresponds to the absorbance of blank

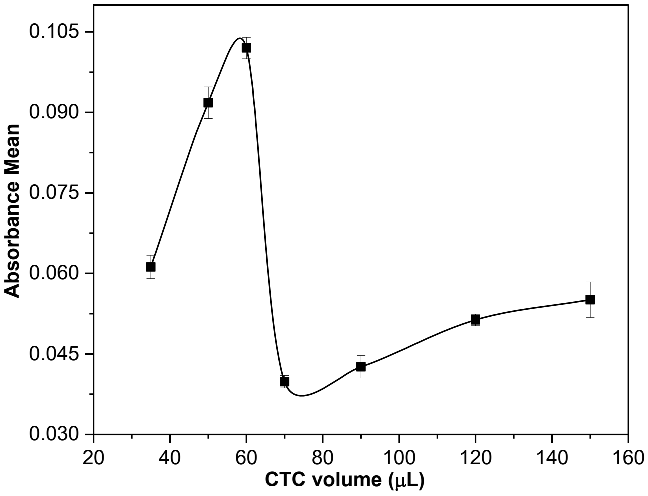

Fig 4. Effect of CTC volume (35–150 µL) on the absorbance of Co(II) using DLLME–FAAS

So that we could select the best disperser solvent for the pre-concentration of Co(II), four solvents (acetone, acetonitrile, ethanol, and methanol) were evaluated in this procedure. An amount of 1000 µL of acetone solvent together with 60 µL of extraction solvent, CTC, and 120 µL of 0.25% (m/v) SAN solution. The experiment was then repeated under the same above conditions with other solvents. These tests revealed that the sedimented phase could be observed with all four disperser solvents but with different signal responses (Fig. 5). From this data, the most obvious finding to emerge from the analysis is that obtained were using ethanol solvent. After that, it became the dispersant for the proposed DLLME procedure.

Usually, the volume of ethanol (dispersing solvent) was tested for the values of 0.50, 0.75, 1.00, 1.25, and 1.50 mL by semi-fixing the CTC solvent volume at 60 µL with ±4 µL. The most striking result to emerge from this test is that the best micro-extraction efficiency of Co(II)–SAN was achieved by using 1000 µL of ethanol. Nevertheless, this experiment did not identify a clear dispersion when using less than 500 µL of ethanol to disperse CTC. A clear increase in absorbance occurred with successive increases in volume of ethanol, from 500 to 1000 µL, while a gradual reduction in microextraction of Co was observed when using more than 1000 µL of ethanol. A possible explanation for this might be a fine droplet of CTC did not form completely, and the extraction is damaging because the solubility of the Co–SAN complex was increased. Thus, the optimal volume for achieving a stable cloudy system is 1000 µL.

Influences of Salting Addition

Experiments were conducted in the absence and in the presence of KNO_3, NaCl, and Na_2SO_4 (31250–187500 m/m) to determine the effect of adding ionic salt on the performance of the DLLME method. Fig. 6 reveals the results obtained from the study of salting out influence in several manners. First, the successive increases of KNO_3 and NaCl concentrations showed no significant

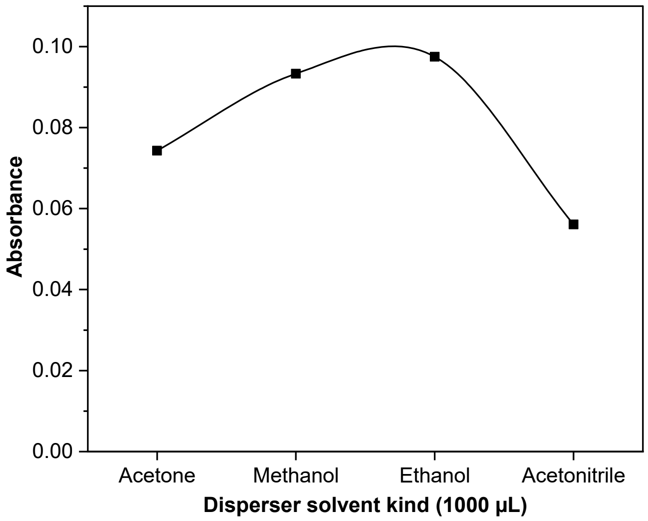

Fig 5. Effect of disperser solvent kind (acetone, acetonitrile, ethanol, and methanol) on absorbance of Co(II) using DLLME–FAAS

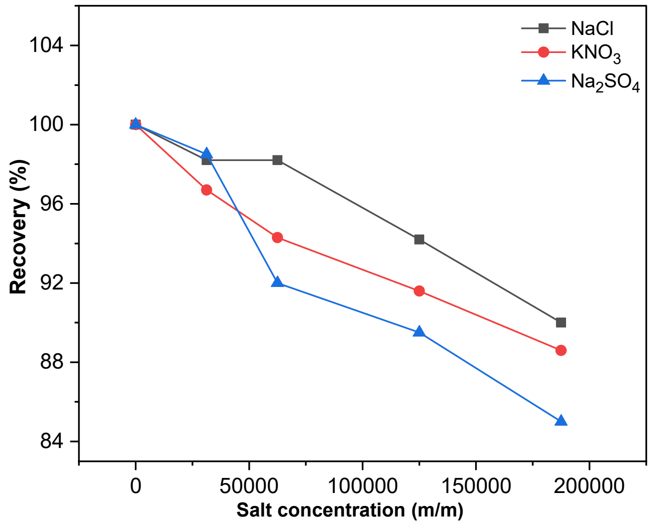

Fig 6. Effect of ionic strength on the absorbance of Co(II) using DLLME–FAAS

effect on the analytical signal. Second, in the high concentration of Na_2SO_4, more than 125000 m/m, there is a slight trend of decreasing transfer of absolute mass of Co(II) ions toward the organic phase. There are several possible explanations for these findings: at high ionic strengths, the possibility that the diffusion rate of Co(II)–SAN complex towards the CTC solvent may be reduced. In addition, the initial charge distribution of amphiphilic molecules is disrupted, hence preventing the formation of coacervate. There are similarities between the results expressed in this proposed work and those described with that of [33] and also consistent with the literature [34-35] conducted without ionic salt.

Performance of FAAS/DLLME Analytical Proposed Method

For the quantification of Co(II) in water, plants, vegetables, and molluscs by FAAS/DLLME, the extract (sedimented organic phase) was nebulized into the bottom directly of the conical tube. The spectrometer was calibrated using aqueous Co standards with concentrations ranging from 4.0 to 160 μg L^{-1} for normal calibration. All of the standards (n = 3) were submitted to the optimized DLLME-Co(II) method, and calibrated extracts were introduced into the FAAS nebulization system in the same manner as sample extracts. In this linear calibration, the instrument measurement parameter, zero intercept, was performed by the intercept with the absorbance signal axis estimated. The results of the analytical curves related to the proposed method, direct measurement of aqueous Co(II) standards, and matrix matching graph are set out in Table 1.

Enrichment factor (EF) is typically determined by dividing the slopes of the analytical standard curves with DLLME procedure/direct measurements [36]. EF results were obtained throughout the work by two methods. First, the slope ratio for DLLME–FAAS vs aqueous standards–FAAS. Second, the ratio for DLLME-FAAS/matrix matched-FAAS. It is found to be 99 and 23. The calculation of the ratio between the aqueous phase volume (10.0 mL) and the final volume of the sediment phase (0.50 mL), this work = 20, is defined as the pre-concentration factor PF. Also calculated is the consumptive index (CI) [36], which represents the volume of sample required to obtain a unit of the EF. The formula CI = V/EF was utilized for the calculations. After putting in the values EFs, 0.43 and 0.10 were the determined CI.

The minimum detectable Co level was estimated by the limit of quantification (LOQ) and limit of detection (LOD). The LOD equation is $3s/m$, and LOQ is $10s/m$, where s is the standard deviation for the 7 replications of the blank samples and m is the slope of the DLLME/FAAS standard curve. Putting in the values $s = 2.31 \times 10^{-4}$ and $m = 6.65362 \times 10^{-4}$ $(\mu g\ L^{-1})^{-1}$ gives LOD = 1.04 μg L^{-1}, and LOQ = 3.47 μg L^{-1}. These minimum detectable Co levels attained by the proposed procedure corresponded to those reported in the previously published research [5,37], and the levels observed in this investigation are far below those observed by Bahar and Babamiri [38] and Mohammadzadeh et al. [39].

Under optimized conditions, precision was estimated using the results of 3 replicate experiments for three levels of Co (low, medium, and high) solution. The consistent results were obtained throughout the work, as shown in Table 2.

Table 1. The analytical characteristics related to the pre-concentration and direct quantification of cobalt

	Aqueous calibration/FAAS	Matrix-matched-calibration/FAAS	DLLME/FAAS-based calibration
Regression equation	Abs = 6.73571×10^{-6} Conc + 0.00132	Abs = 2.89037×10^{-5} Conc + 0.00195	Abs = 6.65362×10^{-4} Conc + 0.00327
Dynamic range (μg L^{-1})	800–3200	80–3200	4.0–160
LOD (μg L^{-1})	112.24	27.50	1.04
EF	99	23	222

Selectivity

Considering the matrix ions effect and absence of selectivity of SAN towards Co ions, those may negatively or positively affect its detection with proposed DLLME–FAAS. Under conditions that have been optimized, the tolerance of the present DLLME method to coexisting some cations was assessed by adding various levels of Al(III), Fe(III), Pb(II), Zn(II), Ca(II), Mg(II), Ni(II), Cu(II), Hg(II), Mn(II), and Cd(II) individually to 50 mL of 80 µg L^{-1} Co(II) standard solution. The results are presented in Table 3. The amounts studied from 11 interfering species yielded a change in cobalt recovery of just over ±14%. The recovery of the Co(II) for the 20% of the species studied declined from 100.0% in the cobalt signal alone to 86%. In general, the DLLME–FAAS method tolerates high levels of potential interfering species.

Accuracy

The evidence for accuracy evaluation was based on the application of this proposed method to different Co(II) levels in environment matrixes (water, plant, vegetables, and molluscs). This evidence has been intimated in two manners: A spiking test was performed by adding a suitable Co(II) amount (2.50 µg L^{-1}) and (4.00–0.50 µg g^{-1}) of Co(II) solution to the real samples. The complete DLLME process was applied after the Co(II) spike, followed by Co(II) quantification by FAAS (Table 4), and the accuracy was evaluated as the recovery ranged from 96 to 107% for a sample with a different

Table 2. The results of the validation experiment

Sample (µg L^{-1})	SD (µg g^{-1})	%RSD
Low Co(II)	0.02	11.7
Medium Co(II)	0.10^{A}	4.7
High Co(II)	0.46	10.2

^{A}µg L^{-1}

Table 3. Results for some matrix cations influence on DLLME efficiency of Co(II)-SAN system with recovery obtained for each proportion

Cations/Co(II) added (m/m)	Recovery %		
	10:1	50:1	150:1
Al(III)	97.0	96.4	93.6
Ca(II)	98.3	94.1	89.7
Cd(II)	94.2	92.9	92.0
Cu(II)	95.1	90.4	85.7
Fe(III)	97.5	93.7	90.5
Hg(II)	99.2	97.8	96.1
Mg(II)	98.9	94.6	94.1
Mn(II)	96.5	94.2	93.8
Pb(II)	98.5	97.0	95.3
Ni(II)	96.1	90.4	89.0
Zn(II)	96.8	90.6	85.5

Table 4. Results (n = 3, ± standard deviation) of recovery experiment for Co(II) quantification in real samples

Sample	Co(II) concentration (µg L^{-1})	Co(II) concentration added (µg L^{-1})	Co(II) concentration found (µg L^{-1})	Recovery (%)
Tap water^{A}	0.72 ± 0.05	2.50	3.14 ± 0.26	97
Tap water^{B}	0.80 ± 0.03	2.50	3.30 ± 0.14	100
Bottled water	N.D	2.50	2.63 ± 0.10	105
Well water^{C}	2.14 ± 0.10	2.50	4.81 ± 0.13	107
River water^{D}	4.01 ± 0.30	2.50	6.65 ± 0.41	106
River water^{E}	4.30 ± 0.38	2.50	6.70 ± 0.60	96
Sample	Co(II) concentration (µg g^{-1})	Co(II) concentration added (µg g^{-1})	Co(II) concentration found (µg g^{-1})	Recovery (%)
Lettuce	4.51 ± 0.46	4.00	8.35 ± 0.72	96
Nigella sativa	0.17 ± 0.02	0.50	0.69 ± 0.04	104
Mentha piperita	2.20 ± 0.24	2.50	4.53 ± 0.41	93
Anethum graveolens	0.85 ± 0.02	2.50	3.48 ± 0.11	105
Bivalve molluscs	1.27 ± 0.104	0.98	2.20 ± 0.15	95

^{A}From treatment and desalination water system of Al-Muthanna, Al-Rumaitha, Iraq. ^{B}From analytical chemistry lab, Al-Muthanna, Iraq. ^{C}Al-Muthanna governorate, Samawah. ^{D}Al-Rumaitha river water, Southern Iraq. ^{E}Euphrates river water, Samawah city, Southern Iraq

Table 5. Results (in μg L^{-1} and μg g^{-1}, ± standard deviation) for accuracy evaluation of Co quantification using direct GFAAS (n = 3), MWD/GFAAS (n = 3), and Proposed DLLME/FAAS

Sample	MWD^F/GFAAS	Proposed DLLME/FAAS	Relative error (%)
Tap water (μg L^{-1})	0.75 ± 0.03	0.72 ± 0.05	−4.17
River water (μg L^{-1})	4.19 ± 0.26	4.01 ± 0.30	−4.54
Bottled water (μg L^{-1})	N.D	N.D	–
Well water (μg L^{-1})	2.15 ± 0.04	2.14 ± 0.10	−0.61
Lettuce (μg g^{-1})	4.34 ± 0.19	4.51 ± 0.46	+3.92
Nigella sativa (μg g^{-1})	0.15 ± 0.03	0.17 ± 0.02	+12.94
Mentha piperita (μg g^{-1})	2.38 ± 0.11	2.20 ± 0.24	−8.20
Anethum graveolens (μg g^{-1})	0.82 ± 0.06	0.85 ± 0.02	+3.53
Bivalve molluscs (μg g^{-1})	1.20 ± 0.12	1.27 ± 0.104	+5.76

^{F}Microwave digestion (MWD) was utilized for mollusc, vegetables and medicinal plant samples

water matrix. The proposed DLLME- FAAS technique accuracy was also estimated as a result of comparison with those obtained using GFAAS. For comparison purposes, water samples that were pretreatment, as was mentioned in the experimental part were analyzed by GFAAS, and the solid samples were analyzed using MWD/GFAAS (Table 5). Samples that were used for accuracy evaluation provided 95–114% recovery during Co quantification by MWD/GFAAS, while in quantification using DLLME/FAAS, it is provided as in Table 4. The t-test showed that the results obtained with the proposed method showed no significant difference at the level of 95%. The agreement ranged from −0.61 to +12.94 for the analyzed samples.

■ CONCLUSION

This research paper was undertaken to design a DLLME procedure to pre-concentrate and separate the Co(II) from aquatic environments, vegetables, medicinal plants, and bivalve molluscs followed by quantification using FAAS. In the devised procedure in which the reagent SAN was successfully utilized as complexing to pre-concentration and extractor for Co(II) by the DLLME method, a univariate strategy for system optimization was established as an accurate quantitative framework for detecting Co(II) in such matrix. It has proven to be a fast, simple, low-cost, and efficient analytical protocol. By comparing with plasma-based detection techniques as well as the literature, which is cited in the results and discussion part, this work has similar analytical characteristics (such as good values of LOD and LOQ), and presents an excellent alternative for the determination of Co(II). Moreover, the DLLME/FAAS procedure has the benefit of minimizing the organic solvent consumption by a few microliters, which results in little waste.

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■ AUTHOR CONTRIBUTIONS

Saher Abdul Reda Ali Alshamkhawy: Supervision, Methodology, Resources, Writing – review & editing. Zaman Sahb Mehdi: Conceptualization, Investigation, Methodology, Formal analysis, Writing – original draft. The whole manuscript was approved by both authors.

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