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The authors declare that there is no conflict of interest regarding the publication of this paper

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All relevant data is included in the paper

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Saher A.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

ORIGINAL STUDY

Univariate Optimization of Dispersive Liquid−Liquid Micro-extraction for Pre-concentration of Lead From Environmental Matrices, Samawah Region Prior to Quantification Using Flame Atomic Absorption Spectroscopy

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Abstract

In this work, a procedure based on dispersive liquid-liquid micro-extraction for lead (Pb) pre-concentration and quantification in an environmental matrix by flame atomic absorption spectroscopy (FAAS) was applied. A case-study approach was chosen to obtain further in-depth information on the Pb levels. The green chemistry principles have been applied for the pretreatment and preparation of real samples by focusing on some features such as the volume of reagents/sample, employ of energy efficient equipment, and production of waste. A univariate strategy was utilized to achieve the optimum extraction conditions. 1750 μL of acetonitrile containing 100 μL of carbon tetrachloride, were rapid injection into: 12.0 mL of a sample solution adjusted the pH at 9.0 and 0.07% (m v^{-1}) Dithizone. The estimated limits of detection (LOD) and quantification (LOQ) under optimum conditions were 2.85 and 9.5 μg L^{-1}, respectively. The enrichment factors EFs were found to be 60.6 (aqueous standards/DLLME-FAAS) and 30.8 (matrix matched/DLLME-FAAS). It was applied for the analyzing eleven environmental samples. The detectable Pb levels for water samples were ranged from 1.87 to 8.11 μg L^{-1}, while in solid samples were ranged 0.39−4.66 μg g^{-1}. The relative standard deviation for three concentrations (low, medium, and high) were found to be 10.64, 6.87, and 2.7, respectively. The presented system was validated by analyzing eleven environmental samples (water, bivalve molluscs, lettuce, and medicinal plants). The results indicated that high recoveries were obtained in the range of 92−104 %. The lead concentration was found in these samples always lower than permissible maximum levels stipulated by Iraqi Standards Specifications 2009 (number 417) for neutral water. The DLLME/FAAS procedure has proven to be a fast, simple, low-cost, and efficient analytical protocol, therefore allowing for employing this protocol in routine analysis of trace metals in such matrixes.

Keywords: Lead, Solvent micro-extraction, Bivalve molluscs, Environmental samples, FAAS

1. Introduction

Lead is non-essential, a trace element, for humans and is among the most common environmental contaminants found in nature [1]. It is heterogeneously distributed in the environment. For example, multiple sources like combustion of fossil fuel and Pb ammunition can increase the risk of Pb exposure in environment [2]. In addition, it is a neurotoxic element and no levels of exposure to Pb is safe to children [3].

Environmental scientists are requesting that analytical chemists investigate the complicated relations between trace metal levels and living organism by focusing current environmental sample research on the detection of trace metal levels in natural water and vegetables [4,5]. Current studies on environmental samples are being directed as regards

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quantifying and distributing the different physicochemical forms of trace elements in bivalve molluscs and water. Molluscs with bivalve structure are perfect sensing organism to monitoring the trace metals amount of aquatic living organisms as an indirect mechanism to sense local environmental modifications. They are now become a widely established tool for monitoring of water pollution, therefore, the field of analyzing molluscs to essential or non-essential metals [6] can substantially contribute to future water quality management [7]. 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 [8].

Usually, the first step in trace levels analysis involves chemical separation of analyte-matrix, followed by advanced instrumental method suited to the detection of trace elements. Nevertheless, after sample enrichment, FAAS is preferred for determining the principal polluting elements (Cd, Cu, Pb, Ni, and Zn) [9,10]. More sensitive and high cost methods as ICP-OES [11], ICP-MS [12], and GFAAS [13] are adopted for direct determination of ultra-trace levels.

Many researchers have highlighted dispersive liquid−liquid micro-extraction (DLLME) for analytes pre-concentration and extraction for subsequent quantification in environmental samples [14−16]. This rapid technique provides also a high recovery factors and simplicity, which is based on dispersive solvent for absolute mass transfer from the aqueous phase, with help complexation agent, to the organic phase [17]. More recent attention has focused on the pre-concentration and detection of lead ions. Da Silva et al. (2024), developed a DLLME method for sample preparation of Pb from petroleum-derived matrices [18]. Furthermore, several recent articles and systematic reviews of liquid-phase extraction have been undertaken for sample preparation of Pb from environmental matrices [19−22].

Considering that DLLME has a great pre-concentration factor, its, therefore, combination with FAAS is an interesting manner for elements quantification. FAAS method was selected because of its low operating cost, simplicity, selectivity, the convenience with a pre-concentration method (DLLME), appropriate for many environmental samples, and It also free from most interferences [23−28].

This work is initially concerned with the focus on using a miniaturized and environmentally friendly solvent micro-extraction methodology, employment of a procedure based on dispersive liquid−liquid micro-extraction for the pre-concentration of Pb and then determination of it by flame atomic absorption spectroscopy. After the optimization of the analytical procedure, the method was applied for the quantification of trace amounts of lead in an environmental matrix (water, bivalve molluscs, lettuce, and medicinal plants) located in the region of Al-Muthanna, Iraq. The greener of pretreatment of the real samples has been performed.

2. Experimental

2.1. Reagents

A standard solution of Pb(II) with a concentration of 1000 μg mL^{-1}, supplied by Sigma-Aldrich (Germany) and it's utilized for freshly dilute and spiking experiments. Sodium sulphate 99.99 % Suparpur (Germany) and potassium acetate, 98.0 % (Sigma Aldrich) for buffer pH range: 4.30−5. 50, 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. Dithizone (DTZ) was used as a chelating agent was kindly provided by Merck (Germany). 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, utilized for solid samples 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.

2.2. 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. 6 kinds of water samples from Al-Muthanna, Southern Iraq, 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 those 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 by purchased from the local market in Samawah, Iraq. The microwave digestion (MWD) procedure for samples 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, 2.0 g of black seed and 400 mg of mint and dill. Each sample was submitted to MWD system. The reagents of digestion process are consists of 6.0 mL of concentrated nitric acid, 2 mL water, and 1.0 mL of 30 % H_2O_2. For spiking experiments of vegetable, an appropriate level of reference cobalt was added to the digested solution and submitted to the DLLME procedure.

2.3. Instrumentation

The instrument employed for spectroscopic measurements of Pb solutions is atomic absorption spectroscopy (AAS) equipped D2-lamp system for background correction (AA-7000 Spectrometer, Shimadzu, Japan) during the quantification of Pb. A single element source was operated at 10 mA. Secondary wavelength, 283.3 nm, was selected for this work, with 0.7 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 15.0 L/min for support gas and 2.0 L/min for fuel gas.

A Hettich centrifuge carried out the physical separation of two phases (model EPA 200, Germany) for the separation of the sedimented phase in DLLME procedure and the undissolved real sample parts. The pHmeter (inoLab pH 7110 xylem analytics, WTW, Germany), connected with an 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 injection of solvents mixture.

Samples were digested by employing a microwave acid (MWD) digestion apparatus with an single reactor chamber (SRC-UltraWAVE, Milestone, Italy). The MWD apparatus is outfitted with an PTFE vessel (capacity of each: 15.0 mL) for fifteen flasks for digestion, as well as a maximal pressure, power, and temperature of 199 bar, 1500 W, and 300 C, respectively.

2.4. Optimization of DLLME

The initial assessment involved examining the production of organic sedimented phase resulting from the combination of three types of extraction solvents: carbon tetrachloride (CTC), chloroform (CHF), and 1,2-dichloroethane (DCE) and disperser solvents (C_3H_6O, C_2H_3N, CH_4O, and C_2H_6O). The experiments were conducted utilizing 1750 μL and 100 μL of disperser and extraction solvents, respectively, along with evaluating the constant volume, 12 mL, of the sample. Subsequently, investigations were conducted the extractor type (CTC, CHF, and DCE) and its volume (50-200 μL), the type and volume (1.0-2.25 mL)of the disperser (C_3H_6O, C_2H_3N, CH_4O, and C_2H_6O).

Additional factors, as sample's pH level (6.0–10.5), DTZ concentration (500 μL of 0.025–0.1 % $m.v^{-1}$), the duration of extraction (0.5–5 min), the impact of including ionic salts (KNO_3, NaCl, and Na_2SO_4) at various amounts, and the duration of centrifugation (ranging from 1 to 10 min).

2.5. Procedure of DLLME

A 12 mL of sample or a solution containing Pb at the μg/L levels and 30 mg mL^{-1} NaCl salt were transferred to a 15 mL test tube (conical bottom), which adjusted the pH value at 9.0. This solution also contained 500 μL of 7.52 % ($m\ v^{-1}$) DTZ. The mixture was gently stirred for 20 s with a vortex mixer to disperse the ligand solvent uniformly throughout the aqueous solution and for accelerate the complex formation. Thereafter, solvents solution: 100 μL carbon tetrachlorid (extraction solvent) and 1750 μL acetonitrile (dispersive solvent) were mixed and rapidly added into the test tube by a syringe. During injection, the micro-droplets formed was dispersed throughout the tube, and the resultant product was left for 1.0 min to complete the extraction. The complex of Pb(II)-DTZ was then extracted in fine droplets of carbon tetrachlorid. Physical separation was achieved by centrifuging the system at 4000 rpm for 4.0 min. Finally, the sedimented organic phase was dilute to 0.50 mL with ethanol to measure the Pb absorbance by FAAS. A scheme for the DLLME procedure is provided in Fig. 1.

3. Results and discussion

The combination of FAAS with DLLME procedures was employed for pre-concentration and quantification of lead ions in environmental matrixes, Samawah region. A univariate strategy for optimization of these procedures from the water, medicinal plant, vegetables, and molluscs samples: DTZ as complexation agent based traditional DLLME.

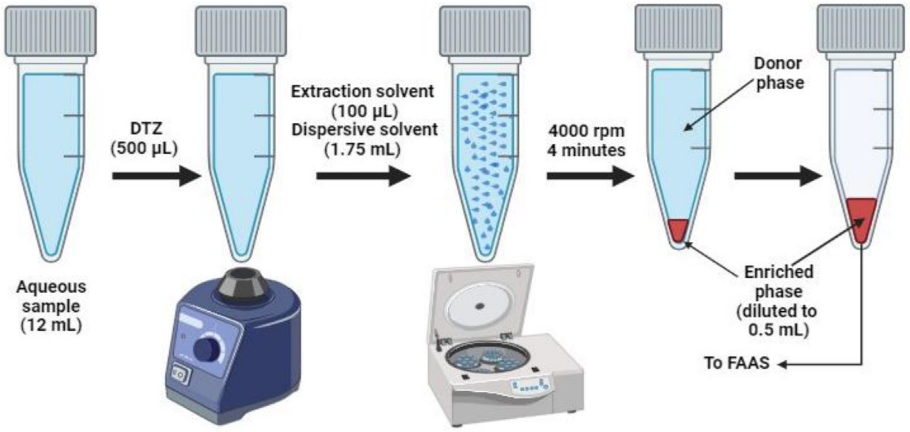

Fig. 1. Schematic of all steps of the dispersive liquid−liquid micro-extraction procedure for lead determination.

3.1. Optimization of complex formation

Influence of pH: When a combination of microwave digestion and DLLME procedures within single strategy, it is essential for achieve a medium of extractor that is compatible with FAAS-DLLME methods [29,30]. The pH of the Pb(II) solution (consisting of 120 μg L^{-1} and 0.03 g/mL NaCl) was investigated from pH 6.0 to 10.5. Several reports [6,31] have shown the optimal ranging to the investigated target was between 5 and 12. Below the pH 6 level, the H_3O^+ in solutions protonates the DTZ (2 nitrogen atoms of ligand) and negatively impacts on complex formation, and a significant amount of lead could be precipitation at higher pH 10 values [32]. Fig. 2 shows an overview of the results obtained from the 6 to 10.5 analysis of pH, the best signal was obtained when employed buffer solutions with a pH in range of 8.5-9.5. From the data in figure below, we can see that the highest signals were recorded at pH 9. The literature has reported the same results for Pb(II) micro-extraction following complexation with DTZ and extraction in a pH 9 medium [31]. Therefore, it requires the pH adjustment at 9.0 for further investigations.

DTZ concentration: The influence of DTZ concentration (0.025, 0.04, 0.06, 0.07, 0.10, 0.5, and 1.0 %) on the values of absorbance of Pb(II) was investigated. The results, as shown in Fig. 3, indicate that best absorbance resulted in DTZ amounts 0.07 %, 0.10 %, and 0.5 % w/v. What stands out in this figure is the absorbance were constant to 0.5 % w/v. Therefore, 0.07 (m v^{-1}) % DTZ solution prepared with an ethanol solvent was selected as best value to further studies.

3.2. Type and volume of extraction solvent

The extraction solvent was chosen based on the highest absorbance (target sample) and the smallest

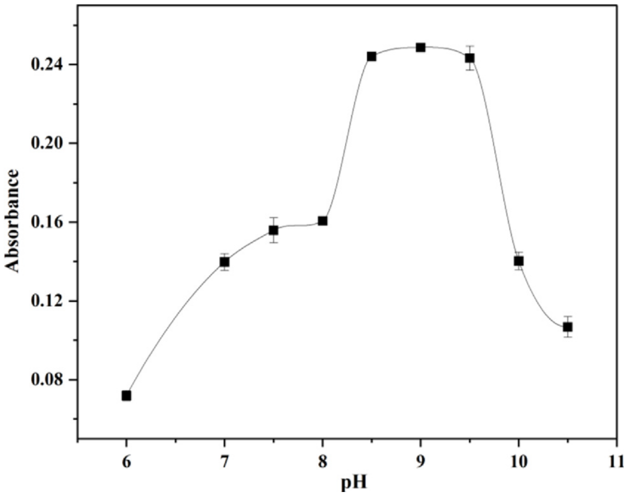

Fig. 2. Effect of pH (6.0−10.5) (n = 3) on absorbance of Pb(II) using DLLME-FAAS.

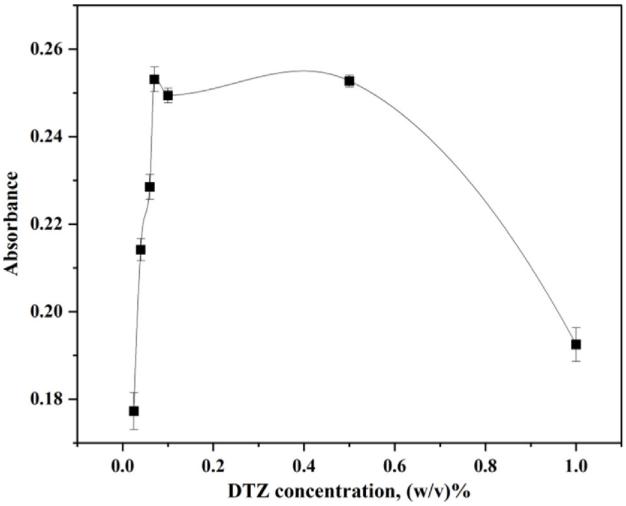

Fig. 3. Effect of DTZ amount on absorbance of Pb(II) using DLLME-FAAS.

blank value. DCE and CTC showed signals with a 10% difference between them, but the blank and % RSD values were lower for CTC (Fig. 4). As mentioned in the literature review, using DTZ as a chelation agent generates a stable Pb(II)-DTZ complex that can be extracted using chlorinated solvents [33,34]. This finding was also reported by Zhou et al. procedure for Pb micro-extraction (DLLME) after complexation using DTZ and extraction using CTC [35]. Therefore, CTC was selected for future investigations.

The volumes of extraction solvent, CTC, is adjusted to achieve maximum DLLME efficiency depending on Pb(II)-DTZ complex while minimizing volume. Procedure for this system was performed using 50, 75, 100, 125, 150, 175, and 200 μL CTC for Pb(II)-DTZ complex extraction. All rich phase is introduced in the nebulization system of spectrometer before being diluted with methanol. This composition of extract gave to good aspiration. In Fig. 5, This study found that the absorbance value is greater when the CTC volume is between 75 and 125 μL and from the graph below we can see that the efficiency of 100 μL of solvent reported significantly more efficiency than other volumes. Consequently, 100.0 μL of CTC was used for subsequent tests.

3.3. Type and volume of disperser solvent

The influence of dispersant solvents type on the DLLME method of Pb(II) was examined utilizing 1750 μL of every type of solvents together with 100 μL CTC, and 500 μL of 0.07% (w v^{-1}) DTZ solution. The result, as presented in Fig. 6, indicate that the best absorbance was obtained when using acetonitrile. After then, this solvent was employed a dispersant for this procedure.

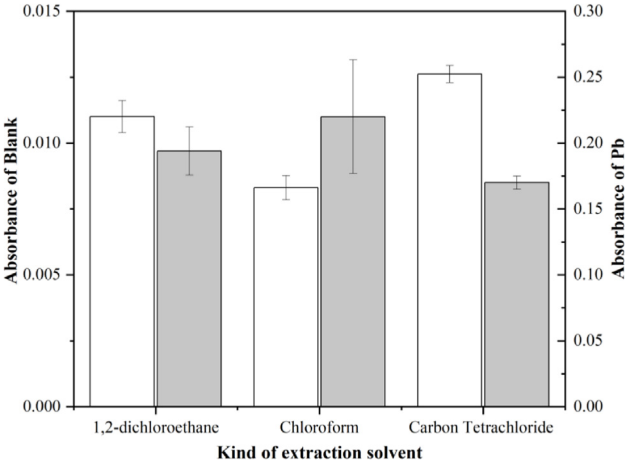

Fig. 4. Effect of kind of extraction solvent (DCE, CHF, CTC) (n = 3) on absorbance of Pb(II) using DLLME-FAAS. (White column/bar corresponds to the absorbance for lead while the grey bar corresponds to the absorbance of blank).

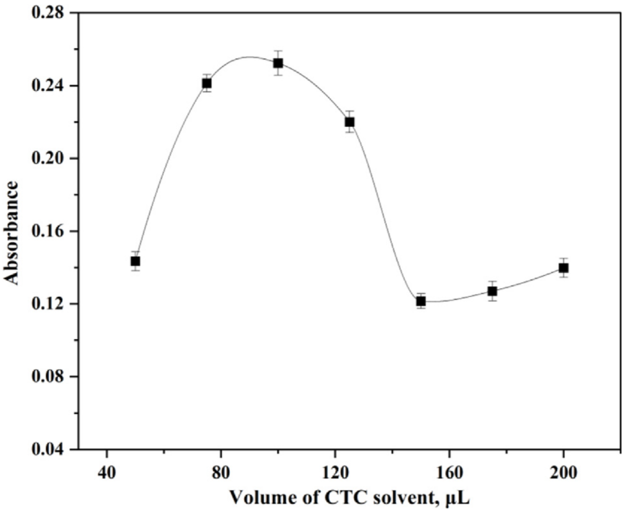

Fig. 5. Effect of CTC volume on the absorbance of Pb(II) using DLLME-FAAS.

The acetonitrile (ACN)volume was tested because of its effect on the efficiency of CTC dispersion and volume of extracted Pb(II)-DTZ. Fig. 7 presents the results obtained from the study of ACN volume effect on the dispersion efficiency. Increasing the acetonitrile to 1.75 mL led to a maximum micro-extraction of Pb(II)-DTZ. A clear increase in absorbance occurred with successive increases in ACN volume, while a gradual reduction in micro-extraction of Pb(II) was observed when using more than 2.0 mL or more of acetonitrile. Thus, the optimal volume for achieving effective dispersion is 1.75 mL.

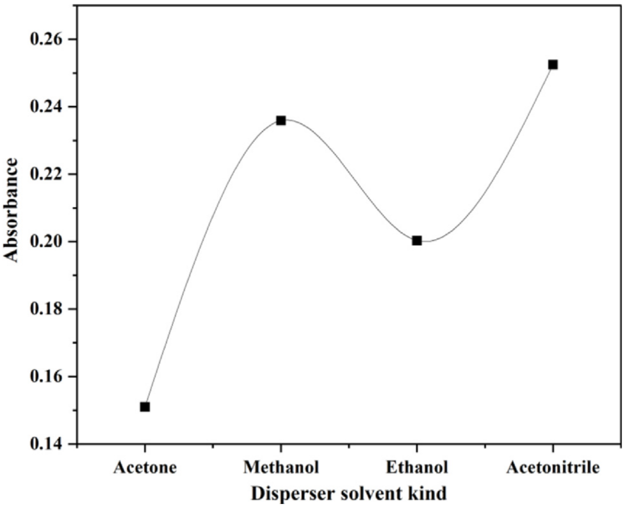

Fig. 6. Effect of disperser solvent kind on absorbance of Pb(II) using DLLME-FAAS.

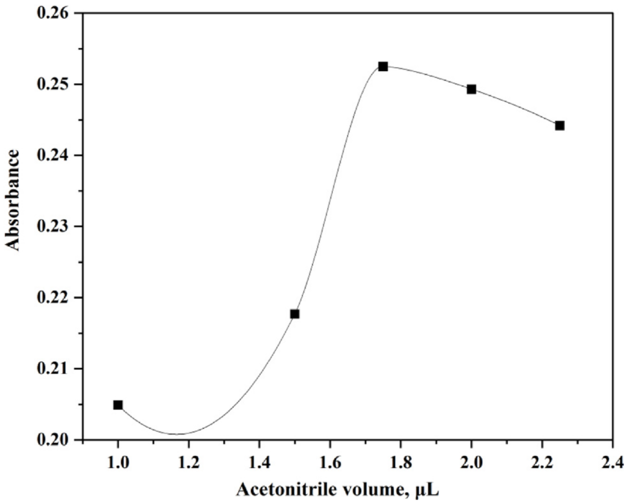

Fig. 7. Effect of ACN volume on absorbance of Pb(II) using DLLME-FAAS.

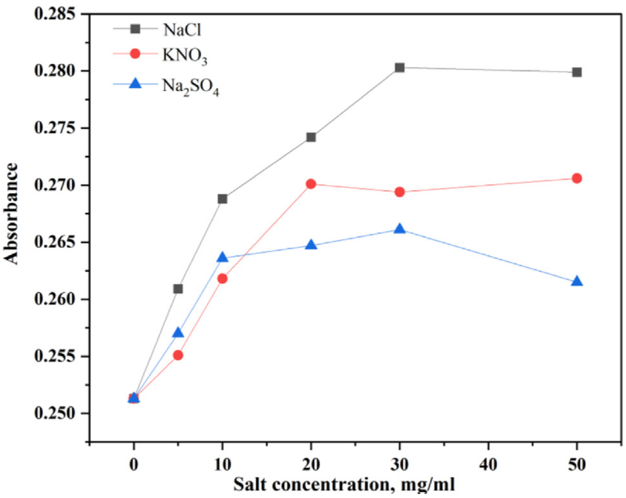

Fig. 8. Effect of adding salt amount (KNO_3, NaCl, and Na_2SO_4) on absorbance of Pb(II) using DLLME-FAAS.

3.4. Influences of ionic strength

Adding ionic salts may increasing the DLLME efficiency by increasing the transfer of an absolute amount of sample target toward the CTC solvent [36]. On the other, a rise in salt concentration may impede the dispersion of the extraction solvent within the target solutions, which could make the extraction less effective [37]. Therefore, to assess the impact of salts addition on the efficacy of the Pb(II)-DTZ-based DLLME technique, a series of experiments was carried out using the specified condition: the inclusion of KNO_3, NaCl, and Na_2SO_4 at concentrations ranging from 0 to 50 mg mL^{-1} into Pb(II)-DTZ complex. The experiments revealed that adding these salts positively impacts the extraction efficiency of Pb(II)-DTZ into the CTC solvent, as depicted in Fig. 8. However, at concentrations between 30.0 mg mL^{-1} and 50.0 mg mL^{-1}, NaCl exhibited a greater absorbance than other salts. Therefore, 30.0 mg mL^{-1} of NaCl was selected and added in this procedure. This result was previously described with the Pb(II)-DTZ-based DLLME method [6] and when DTZ is employed as a ligand-based CPE procedure [38]. This finding is contrary to previous studies which have suggested that the addition of ionic salts reduces the Pb(II)-DTZ extraction efficiency into extraction solvent [39].

3.5. Application and performance of the DLLME/FAAS procedure

The characteristics of analytical curves related to the pre-concentration and direct methods are illustrated in the Table 1: The spectrometer was calibrated using aqueous Pb(II) references with levels that ranged from 10.0 to 360 ng mL^{-1} for external calibrations. All samples target and concentrations prepared have been submitted ($n = 3$, RSD % $= 0.45–6.9$) for the optimized Pb(II)-DLLME method. The extracted of Pb(II) from references and samples has been introduced in the aspiration system of FAAS, by the same manner. The table below illustrates the results of the linearity and its equations, the dynamic ranges at µg L^{-1}, LODs, and EFs analysis for aqueous calibrated standards (0.72, 1.44, 2.88, 4.32, and 8.64 µg mL^{-1}), matrix-matched (0.24, 0.72, 1.44, 2.88, and 8.64 µg mL^{-1}) and DLLME -based calibrated (10, 30, 60, 120, 180, and 360 ng mL^{-1}).

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

	Aqueous calibration/FAAS	Matrix matched- calibration/FAAS	DLLME/FAAS-based calibration
Regression equation	Abs $= 3.86152 \times 10^{-5}$ X $+0.0018$	Abs $= 7.59337 \times 10^{-5}$ X $+8.17 \times 10^{-4}$	Abs $= 0.00234$ X $+ 2.11 \times 10^{-4}$
R-Square	0.9989	0.9993	0.9996
Concen. range, mg L^{-1}	0.72–8.64	0.24–8.64	10-360 µg L^{-1}
LOD, mg L^{-1}	0.083	0.087	2.85 µg L^{-1}
EF	60.6	30.8	139.5

EF results were obtained by two methods. First, the slope ratio for DLLME method/direct method. Second, the slope ratio for Pb(II)-DLLME method/Pb(II)-matrix matched method. It is found to be 60.6 and 30.8. The pre-concentration factor PF = 24 (also, theoretically equal 139.5, as a extracted volume). The consumptive index (CI) was also determined which was found equal 0.2 and 0.39, and 0.09 for all calibration systems.

Minimum detectable Pb(II) level was estimated by LOQ and LOD for the 7 replicate of the blank DLLME experiment by putting in the values $s = 2.22 \times 10^{-3}$ and $m = 2.34 \times 10^{-4}$ $(\text{ng mL})^{-1}$gives LOD = 2.85 ng mL^{-1}, and LOQ = 9.5 ng mL^{-1}. This minimum detectable Pb(II) levels attained by this system are in line with those of recent studies [40] However, this finding is contrary to previous studies which found that the LOD = 7 μg L^{-1} [41] and study by [42] as 11.4 μg L^{-1}, while the LOD levels observed in this investigation are above from those observed by Parween et al. and Gholmi et al. [43,44] respectively.

3.5.1. Precision

Under optimized conditions, precision was estimated using the findings of 3 replicate experiments for low, medium, and high Pb(II) solution levels. From data in Table 2, the consistent results were obtained all throughout the work. In addition, we can see that analysis *A. graveolens* for Pb resulted in the lowest value of RSD.

Table 2. The results of validation experiment

Sample,	±SD	%RSD
1.87 μg L^{-1}	0.2	10.64
0.74 μg g^{-1}	0.02	2.7
4.66 μg g^{-1}	0.32	6.87

3.5.2. Selectivity

Considering the matrix ions effect and absence of selectivity of DTZ towards Pb(II)s those may negatively or positively affect on target detection with DLLME-FAAS. Under conditions that have been optimized, the tolerance of present DLLME for coexisting cations has been assessed with present interfering ion as can be seen from the table below, at various levels to Pb(II) standard, the results are presented in Table 3.

Having into consideration which a significant interfering happens when the potential interference ion alone causes an shift in Pb recovery of ±10.0 %, it is noticed that the Pb(II)-DTZ based-DLLME has,

Table 3. Results of the influence of some matrix cations on DLLME efficiency of Pb(II)-DTZ system (120 μg L^{-1} Pb solution) with % recovery obtained for each proportion

Coexisting ions	Proportion of coexisting ions/lead ions		
	1:1	10:1	50:1
	(Recovery %)		
Al^{+3}	99	97.5	91.2
Ca^{+2}	100.9	101.6	102.9
Co^{+2}	98.3	98.1	92.7
Cu^{+2}	99.1	96.5	93.4
Fe^{+2}	98.6	95.8	90.1
Mg^{+2}	99.2	99	97.8
Mn^{+2}	99.5	99.1	97.3
Cd^{2+}	102.6	104.3	104
Ni^{2+}	99.3	99.7	108.2
Zn^{2+}	98.8	97.5	94.5

Table 4. Results (in μg/L and μg/g, n = 3, ±standard deviation) of recovery experiment for Pb quantification in real samples.

Sample	Pb concentration in original sample (μg/L)	Pb concentration added (μg/L)	Pb concentration found (μg/L)	Recovery (%)
Tap water[a]	6.32 ± 0.09	25.0	29.86 ± 1.06	94
Tap water[b]	8.11 ± 0.23	25.0	33.21 ± 0.19	101
Bottled water	N.D	25.0	25.2 ± 2.21	101
Well water[c]	N.D	25.0	24.78 ± 0.9	99
River water[d]	1.87 ± 0.2	25.0	27.15 ± 3.27	101
River water[e]	3.74 ± 0.18	25.0	28.64 ± 0.75	100
Sample	Pb concentration (μg/g)	Pb concentration added (μg/g)	Pb concentration found (μg/g)	Recovery (%)
Lettuce	N.D	2.00	2.03 ± 0.16	102
Nigella sativa	N.D	0.50	0.46 ± 0.08	92
Mentha piperita	0.39 ± 0.03	1.25	1.69 ± 0.2	104
Anethum graveolens	0.74 ± 0.02	1.25	1.91 ± 0.25	94
Bivalve molluscs	4.66 ± 0.32	1.96	6.58 ± 0.13	98

[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.

Tables 5. Comparison of the proposed system with different other procedures for the pre-concentration and quantification of lead.

Method	matrix	Detector	DR, μg L^{-1}	LOD, μgL^{-1}	PF	V, mL	Reference
DSPME	Water and tea	FAAS	60–600	8.9	15	15	[45]
DLLME	Water	FAAS	–	4.3	50	10	[46]
DLLME	Molluscs and sea water	FAAS	10–100	12	32/31.7	8.0	[6]
SPE	water and pharmaceutical products	ICP-OES	10–80	6.36	20	20	[47]
SUPRAS-SME	Herb plants and Wastewater	FAAS	15.1–606	0.69	33	10	[19]
DLLME	Water	FAAS	1–70	0.5	10^3/450	25	[48]
Coprecipitation	Water	FAAS	–	16	125	50	[49]
CPE	Tap and mineral water, crushed wheat, and tomato paste	FAAS	0.5–1.5 2.0–12	7.2	10	10	[50]
DLLME	Water, Molluscs, plants, and vegetables	FAAS	10–360	2.85	140 30.8 60.6[a]	10	Present method

[a] EF is calculated as the ratio of slope of preconcentrated samples to that obtained without pre-concentration (aqueous standard calibration).

in general, a satisfactory level of tolerance towards potential interference ion. These results demonstrate that this method can be applied onto real specimens in various environmental matrixes.

3.5.3. Accuracy

The evidence for accuracy evaluation was based on application of this proposed method to different Pb(II) levels in environment matrixes (water, plant, and mollusk). This evidence has been intimated by a spiking test which was performed by adding a suitable Pb amount (25.0 ng mL^{-1} lead) to the real sample. The complete dispersive liquid-liquid micro-extraction process was applied after the Pb spike, followed by quantification of Pb by FAAS (Table 4), the accuracy was evaluated as the recovery (R) ranged from 92 to 104 % for a sample with a different water matrix.

4. Conclusions

This work was undertaken to employ a modern pre-concentration procedure (DLLME) to sample preparation of Pb from aquatic environments, vegetables, medicinal plants and bivalve molluscs and then quantification using FAAS. A univariate strategy for system optimization was established an accurate quantitative framework for detecting of target analyte in such matrix.

The results of these investigations show that the enhancement power of Pb quantification has been improved by 139.5 times compared to the results of FAAS alone. Furthermore, by compared with plasma-based detection techniques as well as the literature which is cited in the Table 5 and in results and discussion part, this work has similar analytical characteristics (such as good values of LOD), and provides a good alternate for the Pb quantification. Comparable results from high sensitive technique and quantitative recoveries have been obtained, with low consumption of solvents and does not require the employment of large volumes from highly dangerous chemicals required for pretreatment (as, HNO_3).

It is important to mention that the concentrations of Pb in bivalve (fresh water) and water did not follow the guidelines established by international authorization (US). In addition, the lead concentration was found in these samples always lower than permissible maximum levels stipulated by Iraqi Standards Specifications 2009 (number 417) for neutral water. Moreover, the study's findings demonstrated that the suggested approach exhibited a high tolerance for interfering ions.

The DLLME/FAAS procedure has proven to be a fast, simple, low-cost, and efficient analytical protocol. Moreover, the proposed methods presents an excellent alternative for the determination of the target element, with similar characteristics to the existing methods in addition to requiring a low volume of samples and these methods also have the benefit of minimizing the organic solvent consumption by a few microliters, which results in little waste. Finally, the low cost, simplicity, good sensitivity, and lower extraction time allowed for a capable to employ this protocol in routine analysis of trace metals in such matrixes.

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