Synthesis and identification of meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone (MBBOTSC) as novel ligand for cadmium extraction by ultrasound assisted-dispersive-ionic liquid-liquid microextraction method

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\textbf{ABSTRACT}

In this research, meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone (MBBOTSC) as a novel ligand was synthesized from the reaction between meta-(4-bromobenzyloxy) benzaldehyde and thiosemicarbazide under basic condition in water and ethanol as solvents. Ligand has the ability to chelate ions and therefore, it was used to form a complex and extract ions. So, the cadmium ions in water and wastewater samples were separated based on MBBOTSC by ultrasound assisted-dispersive-ionic liquid-liquid microextraction method (USA-D-ILLME) before determination by AT-F-AAS. The MBBOTSC ligand was added to the mixture of the ionic liquid/acetone (IL/AC, [OMIM][PF\textsubscript{6}]) and then injected by syringe to 50 mL of water samples at pH 6-7. The sample was put into the ultrasonic accessory for 5 minutes, after complexation (Ligand-Cd; RS…Cd….RS), the water sample was centrifuged for 3 min for phase separation. Due to complexation and back-extraction of Cd in liquid phase, the amount of Cd ions in the water samples was determined by AT-F-AAS after dilution eluent (0.5 M, HNO\textsubscript{3}) with DW up to 1 mL. In optimized conditions, the Linear ranges and LOD for 50 mL of water samples were obtained 1-36\textmu g L\textsuperscript{-1} and 0.3\textmu g L\textsuperscript{-1}, respectively (Mean RSD= 1.26%). The validation results were successfully achieved by spiking real samples and using electrothermalatomic absorption spectrometry (ET-AAS).

\textbf{Keywords:}

Cadmium, Meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone, Ligand, Synthesis, Metal extraction, Ultrasound assisted-dispersive-ionic liquid-liquid microextraction

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\textbf{1. Introduction}

Thiosemicarbazones (TSCs) are an important class of organic compounds containing N and S elements that have various uses and properties [1]. These compounds as Schiff base ligands have been widely used to form complexes and to separate metals [2]. Furthermore, due to the presence of electron pairs on sulfur and nitrogen, these compounds are used over a large area (Fig. 1). TSCs form a prominent class of pharmaceuticals and biologically active compounds by virtue of their antimicrobial, antiviral, anti-bovine viral diarrhea, antiproliferative and antifungal activities [3-7]. Also, the formation of metal complexes plays a very important role in the treatment of cancer, especially since there are reports of the destructive role of reactive oxygen species in increasing the antiproliferative activity of chelators against...
tumour cells [8]. Synthesis of TSC is performed in different ways: a) it is a two-step pathway in which hydrazine reacts with isothiocyanate and the product with aldehyde or ketone, which leads to TSCs [9], b) it is the opposite of pathway a, ie hydrazine first reacts with aldehydes or ketones and then the resulting compound reacts with isothiocyanate [10], c) it is a 4-step process, in which hydrazine first reacts with carbon disulfide and the resulting intermediate reacts with methyl iodide to form methylhydrazine thiocarbamate, eventually nucleophilic substitution with amines and a condensation reaction with an aldehyde or ketone produces TSC [11]. In continuation of our research on the synthesis of various compounds [12-15] and recent report introducing penicillamine as a metal chelator [16], herein, MBBOTSC as ligand containing sulfur and nitrogen has proposed as cadmium chelator.

The toxicity of cadmium (Cd) in water is very important because of the long half-life in the human body. The high toxicity of cadmium caused to damage human organs such as kidneys, liver, lungs and cancer [21,22]. Cadmium is easily transferred from water to plants/humans and has also contamination in the environment [23]. The environmental protection agency (EPA) and Agency for Toxic Substances and Disease Registry reported that cadmium values in the waters are usually less than 5 μg L⁻¹ [24]. For healthy people, the mean cadmium concentration in human serum samples is less than 0.2 μg L⁻¹ [25]. So, the determination of cadmium in water samples is an important concern for controlling toxicity [26,27]. The sensitive techniques such as the electrothermal atomic absorption spectrometry (ET-AAS) [28], the atom trap assist to flame atomic absorption spectrometry and flame atomic absorption spectrometry (AT-F-AAS, F-AAS) [29], inductively coupled plasma–atomic emission spectrometry (ICP-AES) using thiosemicarbazide derivative on alumina [30], and inductively coupled plasma–mass spectrometry (ICP-MS) [31], was used for the determination of cadmium in water and wastewater samples. However, as a low concentration of cadmium and high interferences in wastewater samples, sample preparation before analysis is required. Solid-phase extraction (SPE) and liquid-liquid micro extraction (LLME) are preferred to other techniques for ultra-trace cadmium determination and separation in water samples [32,33].

![An anticancer active thiosemicarbazone](image1)

An anticancer active thiosemicarbazone [17]

![Pd thiosemicarbazone Complex, as Catalysts for Cross-Coupling Reactions](image2)

Pd thiosemicarbazone Complex, as Catalysts for Cross-Coupling Reactions [18]

![Mn(II) thiosemicarbazide complexe as electrochemical oxygenreductive catalyst](image3)

Mn(II) thiosemicarbazide complexe as electrochemical oxygenreductive catalyst [19]

![Isatin thiosemicarbazone as anti-corrosive agent for the alloy](image4)

Isatin thiosemicarbazone as anti-corrosive agent for the alloy [20]

Fig. 1. Several TSC derivatives with different application
Herein a TSC as ligand containing sulfur and nitrogen was synthesized in two steps; a) synthesis of a primary aldehyde; meta-(4-bromobenzyloxy) benzaldehyde, b) synthesis of final ligand meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone (Scheme 1). Then the structure of the final ligand was identified by spectroscopic methods such as NNR and IR as well as TLC and melting point.

In this study, the ligand structure was identified by spectroscopic methods such as 1HNMR, 13CNMR, and FTIR as well as melting point. Thin layer chromatography (TLC) was used to check the progress of the reaction (ethyl acetate and n-hexane were used as the mobile phase). Finally, the mixture of MBBOTSC ligand, IL and acetone was used for cadmium extraction based on the USA-D-ILLME procedure at pH 6-7 before determined by the AT-F-AAS. The results were validated by spiking of water samples and compared to the ET-AAS.

2. Experimental
2.1. Instrumental Analysis
The atom trap flame atomic absorption spectrometry (AT-F-AAS) was used for cadmium determination in water and wastewater samples. The signal absorption improved by the atom trap accessory was added to the burner AAS which was caused to increase the sensitivity of absorption for cobalt analysis. The limits of detection (LOD) for AT-FAAS and FAAS were achieved 0.05 and 0.2 mg L⁻¹, respectively. The information of cobalt was entered to software Avanta (wavelength of 228.8 nm, 3.0 mA and slit of 0.5 nm). The auto-sampler was used for all samples. The linear range for AT-FAAS was 0.05-1.8 mg L⁻¹ for cobalt analysis. The validation of results was checked by electrothermal atomic absorption spectrophotometer (GBC, Aus) in water samples (0.2-6.0 μg L⁻¹). The pH of the water samples was tuned by the buffer solutions of Merck, Germany before measured by the digital pH meter (Metrohm, Swiss). The phosphate buffers have adjusted the pH for 6-7.

2.2. Reagents and Materials
All reagents/starting materials and solvents used in this study were purchased from commercial suppliers (Merck AG, Aldrich or Acros Organics). The melting point was measured on an electrothermal 1A9100 melting point apparatus fixed at 1°C per minute and are uncorrected. The reagents of phosphate buffers (CAS N: 7558-79-4) were prepared from Sigma Aldrich, Germany. The ionic liquid of 1-methyl-3-octylimidazolium hexafluorophosphate ([OMIM] [PF₆], CAS N: 304680-36-2) was prepared from Sigma Aldrich, Germany. The calibration standards containing 1, 5, 10, 20, 25, 35 μg mL⁻¹ Cd standard solutions were prepared by dissolving 1.0 g of cadmium nitrate (Cd(NO₃)₂) in 1 L of deionized water solution (DW in 2% HNO₃). The standard of Cd solutions was daily made by stock solutions (1g L⁻¹, 1000 mg L⁻¹) which was diluted by DW up to one litre (Millipore, USA). Other reagents such as HCl, HNO₃ and acetone were purchased from Merck, Germany.

2.3. Characterization
IR spectra were recorded using Nicolet FT-IR Magna 550 spectrophotographs (KBr disks). The NMR spectra were recorded on Bruker 300 spectrometers in DMSO-d₆ as solvent. 1H NMR data are reported in the following order: Chemical shifts (δ) in part per million (ppm) downfield from TMS as internal standard; approximate coupling constant values (J) in Hertz (HZ); spin multiplicities (s, singlet; d, doublet; t, triplet. Thin-layer chromatography (TLC) was performed on pre-coated Silica Gel F254 plates in ethyl acetate: n-hexane as the mobile phase, for checking the reactions.

2.4. Synthesis of meta-(4-bromobenzyloxy) benzaldehyde
The meta-(4-bromobenzyloxy) benzaldehyde was prepared by reaction of 3-hydroxybenzaldehyde with 4- bromobenzylchloride in the presence of K₂CO₃ as catalyst. The physical and chemical properties of it, was compared with literature and its structure was confirmed [34]
2.5. Synthesis of MBBOTSC

In a 50 ml round bottom flask equipped with a magnet, dissolved 0.1 g of NaOH in 5 cc of distilled water, at first 1 mmol (0.291 mg) of meta-(4-bromobenzyloxy) benzaldehyde and then gradually 1 mmol (0.091 g) of thiosemicarbazide was added to the resulting solution. And the mixture was gently stirred for one day. Then 5 cc of ethanol was added and refluxed for one hour. The reaction progress was controlled by TLC EtOAc/n-hexane (1:3) as eluent. The obtained sediments were filtered using filter paper and dried in a vacuum. Then the precipitate was recrystallized with IL [OMIM][PF₆] at the bottom of a PVC conical tube by centrifuging for 3 min and speed of 4000 rpm. The water samples were set aside by auto-sampler (50 mL) and the Cd ions in IL phase back-extracted by 0.5 mL of HNO₃ (0.5M). After diluted acide phase with DW up to 1.0 mL, the Cd was complexed with the MBBOTSC ligand [R-N: (S:) ……Cd(II)←(:S)N:-R] at pH 6. After the complexation, the Cd-MBBOTSC was separated and nitrogen groups of MBBOTSC ligand [R-Br] at pH 6. After the complexation, the Cd-MBBOTSC was separated with IL [OMIM][PF₆] at the bottom of a PVC conical tube by centrifuging for 3 min and speed of 4000 rpm. The water samples were set aside by auto-sampler (50 mL) and the Cd ions in IL phase back-extracted by 0.5 mL of HNO₃ (0.5M). After diluted acide phase with DW up to 1.0 mL, the Cd values was determined by AT-FAAS. The procedure was shown in Figure 2. The enrichment factor (EF) based on the slope of calibration curve of Cd in the proposed procedure (USA-D-ILLME) and standard method was calculated (m₁/m₂ =tgα, EF= 49.64).

2.6. Procedure of metal extraction

By the USA-D-ILLME procedure, the cadmium was complexed with the MBBOTSC ligand in water and wastewater samples. Also, the Co ions was determined by AT-F--AAS. The MBBOTSC ligand (0.1 g) added to mixture of [OMIM][PF₆] and acetone (0.2 g: 500 μL) and was injected into 50 mL of Cd included 1-36 μg L⁻¹. After shaking of samples (5.0 min), the cobalt ions were extracted with sulfur and nitrogen groups of MBBOTSC ligand [R-Br] at pH 6. The procedure was shown in Figure 2. The enrichment factor (EF) was calculated (m₁/m₂ =tgα, EF= 49.64).

\[
\text{m}_1 / \text{m}_2 = \tan \alpha, \quad EF = 49.64
\]

<table>
<thead>
<tr>
<th>Entry</th>
<th>Structure</th>
<th>Yield (%)</th>
<th>m.p. (°C)</th>
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<tr>
<td>2</td>
<td>![Structure Image]</td>
<td>87</td>
<td>187-189</td>
</tr>
</tbody>
</table>
The aim of the present study was to synthesize the meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone derivative and to confirm its structure by spectroscopic methods such as FT-IR, 1H-NMR, and 13C-NMR and use it to separate and extract the cadmium ions from the water and wastewater samples before determined by AT-F-AAS.

The meta- (4-bromo benzyloxy) benzaldehyde reacted with thiosemicarbazide under basic condition to form the target ligand. Ligand synthesis is based on a nucleophilic attack of the electron pair of the free amine of thiosemicarbazide on the carbon of the carbonyl of aldehyde group, and after removing of a water molecule from intermediate, an imine bond is formed and the product is obtained.

3. Results and Discussion

The aim of the present study was to synthesize the meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone derivative and to confirm its structure by spectroscopic methods such as FT-IR, 1H-NMR, and 13C-NMR and use it to separate and extract the cadmium ions from the water and wastewater samples before determined by AT-F-AAS. The meta- (4-bromo benzyloxy) benzaldehyde reacted with thiosemicarbazide under basic condition to form the target ligand. Ligand synthesis is based on a nucleophilic attack of the electron pair of the free amine of thiosemicarbazide on the carbon of the carbonyl of aldehyde group, and after removing of a water molecule from intermediate, an imine bond is formed and the product is obtained.

3.1. FTIR Analysis

The FT-IR, 1H-NMR, 13C-NMR spectra of the synthesized ligand analyzed to confirm its structure. In the FT-IR spectrum of the ligand, two stretching vibration frequencies in the regions of 3393 and 3342 cm⁻¹ belong to the NH₂ group. The peak that appearing at the stretching vibration frequency of 3025 cm⁻¹ is related to the aromatic CH stretching vibrations of this synthesized ligand. The peak that has appeared at 1530 cm⁻¹ is related to the stretching vibration frequency of the C = N. And the peak appearing at the frequency 1262 cm⁻¹ is correspond to the stretching vibration frequency of the C = S functional group. The peak at 834 cm⁻¹ is attributed to the C-Br stretching vibration (Fig.3).

![Fig.2. Determination of cadmium based on MBBOTSC ligand by the USA-D-ILLME procedure](image)

![Fig.3. FT-IR spectrum of MBBOTSC ligand](image)
3.2. \( ^1H \)-NMR spectrum

In the \( ^1H \)-NMR spectrum of the ligand, a single peak appeared in the region of 11.48 μg mL\(^{-1}\), which corresponds to hydrogen of NH. Two single peaks in the regions of 8.27 μg mL\(^{-1}\) and 8.1 μg mL\(^{-1}\) belong to the two protons NH\(_2\), and the single peak appear at the 8.04 μg mL\(^{-1}\) indicate the CH alkene group. The three aromatic protons appeared as a doublet at the 7.5 μg mL\(^{-1}\) with coupling constant (\(J\)) 6 Hz. In the area of 7.45 μg mL\(^{-1}\), a doublet with coupling constant (\(J\)) 6 Hz appeared, which corresponds to two aromatic protons, as well as in the area of 7.35 μg mL\(^{-1}\), doublet with coupling constant (\(J\)) 6 Hz has appeared related to two aromatic protons. In the region 7.03 μg mL\(^{-1}\), a triplet peak has appeared with coupling constant (\(J\)) 6 Hz, which is corresponded to an aromatic proton. The single peak that appears in the region of 5.15 μg mL\(^{-1}\) belongs to the two protons of the OCH\(_2\) group. The \( ^13C \)-NMR spectrum of this compound also confirms the desired structure in that the ligand contains 11 types of carbon, which can be seen in the \( ^13C \)-NMR spectrum as 11 separate carbon peaks (Fig.4 and 5).

**Representative spectral data**

Meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone (X): Cream-shaped crystals, \textit{m.p.:} 187-189° C; FT-IR (KBr) (\(\nu_{max}\), cm\(^{-1}\)): 3393 (N-H), 3342 (N-H), 3160 (N-H), 3025 (C-HAr), 1530 (C=N), 1262 (C=S), 1170 (C-O), and 834 (C-Br). \( ^1H \) NMR (DMSO-d\(_6\), 300 MHz) \(\delta\) (ppm): 11.48 (1H, s, NH), 8.27 (1H, s, NH), 8.1 (1H, s, NH), 8.04 (1H, s, CH), 7.5 (3H, d, \(J = 6\) Hz, CH\(_{Ar}\)), 7.45 (2H, d, \(J = 6\) Hz, CH\(_{Ar}\)), 7.35 (2H, d, \(J = 6\) Hz, CH\(_{Ar}\)), 7.03 (1H, t, \(J = 6\) Hz CH\(_{Ar}\)), and 5.15 (2H, s, OCH\(_2\)). \( ^1C \) NMR (DMSO-d\(_6\), 75 MHz) \(\delta\) (ppm): 178, 158, 142, 136, 136, 131, 130, 121, 117, 112, and 69.
3.3. Mechanism of cobalt extraction
The mechanism of extraction is based on the interaction of imine nitrogen (=N) and thio (=S) groups of the MBBOTSC with cadmium ions using dative/covalent bonding to form stable five ring. The mechanism of extraction between nitrogen and thiol of the MBBOTSC with Cd ions was shown in Figure 6. The mechanism demonstrates that MBBOTSC ligand can bind to Cd$^{2+}$ by complex formations that are easily eliminated from the water samples. The sulfur and nitrogen groups in MBBOTSC ligands caused to easily extracted the Cd ions from aqueous solutions

3.4. Optimizing of parameters
3.4.1. The effect of pH
The pH of the sample is the main parameter for cadmium extraction from water and wastewater samples which was affected oncomplexation. So, the effect of pH on extraction efficiency of cadmium with MBBOTSC ligand was studied in different pH between 2 to 11 (Fig.7). The complexation of sulfur and nitrogen groups with Cd$^{2+}$ was depended on the pH of samples. Due to results, the efficient extraction for Cd ions was obtained more than 96% at pH of 6.0-7.0 (Cd→:S-R or :NH$_2$-R). At higher pH (more than 7.5), the extraction of cadmium was reduced and Cd ions was precipitated [Cd(OH)$_2$]. The mechanism was depended on the coordination bond between Cd$^{2+}$ and amine /sulfur groups of MBBOTSC ligand at optimized pH. In acidic pH, the NH$_2$ and SH groups of the MBBOTSC ligand are protonated (+) and complexation decreased. Also, in basic pH at more than 7.5, the NH$_2$ and SH groups were deprotonated (-) but the cadmium ions precipitated. So, the MBBOTSC is favorite ligand for extraction and determination of cadmium in water samples by the USA-D-ILLME procedure at pH 6.5.
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3.4.2. The effect of IL

The different hydrophobic ionic liquid such as, [OMIM][PF$_6$], [BMIM][PF$_6$] and [EMIM][PF$_6$] was used for collecting and separation ligand/Cd from water samples (Fig. 8). So, the effect of ILs on the cadmium extraction was evaluated within the IL range (0.1-0.5 g) by standard cadmium solution (1-36 μg L$^{-1}$). Due to results, the high recoveries were achieved by 0.17 g of [OMIM][PF$_6$] (98.9%). Therefore, 0.2 g of [OMIM][PF$_6$] was used as optimum IL for Cd extraction by MBBOTSC ligand.

3.4.3. Optimization of MBBOTSC ligand

The effect of MBBOTSC for cadmium extraction must be evaluated. MBBOTSC is one of the important factors for cadmium extraction which should be optimized by the USA-D-ILLME procedure. First, $0.015 \times 10^{-4} - 0.35 \times 10^{-4}$ mol L$^{-1}$ of MBBOTSC ligand was examined for cadmium extraction in the water and standard samples. Due to results, the recovery of extraction increased for $0.065 \times 10^{-4} - 0.35 \times 10^{-4}$ mol L$^{-1}$ of MBBOTSC ligand (Fig. 9). So, $0.07 \times 10^{-4}$ mol L$^{-1}$ of MBBOTSC ligand in 50 mL of water sample was selected as optimum amount of ligand for cadmium extraction.

3.4.4. Effect of back-extraction eluents

The effect of back-extraction eluents for cadmium extraction in water samples were studied by MBBOTSC ligand. At acidic pH (pH<4), the covalent bond between the cadmium ions and sulfur/nitrogen groups (complexation) was broken and cadmium ions released into eluents. For this purpose, the different acid solutions (HCl, HNO$_3$, H$_2$CO$_3$, H$_2$SO$_4$) were prepared and used for back-extraction cadmium ions from ligand/IL in water and wastewater samples. In addition, the eluent concentrations and volumes between 0.1-1.0 mol L$^{-1}$ and 0.1-2.0 mL) was studied. The results showed, the high extraction for cadmium was achieved by 0.5 mL of nitric acid (0.5 M) (Fig. 10).

![Fig. 8. The effect of amount of IL on cadmium extraction based on MBBOTSC ligand by the USA-D-ILLME procedure](image-url)
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3.4.5. Effect of sample volume
The effect of sample volume for cadmium extraction in water samples was evaluated between 5 - 100 mL with cadmium concentration (1-36 μg L⁻¹) by AT-F-AAS. As result, the high recovery occurred less than 60 mL of water samples at pH 6.5. So, 50 mL was selected as optimum sample volume for cadmium extraction based on the MBBOTSC ligand by the USA-D-ILLME procedure.

3.5. Validation of methodology
The cadmium ions (Cd²⁺) was separated and determined in water and standard samples by the USA-D-ILLME procedure. The cadmium ions were successfully extracted based on MBBOTSC ligand in water samples with high recovery. Moreover, the accuracy of cadmium analysis must be validated by advanced analytical techniques and spiking samples. In this study, the results of USA-D-ILLME procedure was validated by spiking the standard cadmium solution in water samples (Table 2). Also, the data analysis of cadmium in this procedure can be validated by ET-AAS (Table 3). The results demonstrated the accurate extraction and high recovery for cadmium ions in water and wastewater samples. The spiked samples showed satisfactory results for extraction and separation of cadmium based on the MBBOTSC ligand in water samples by the USA-D-ILLME procedure.

4. Conclusions
An analytical method for extraction and determination of cadmium in water samples was carried out by synthesis of meta-(4-bromobenzyloxy) benzaldehyde thiosemicarbazone (MBBOTSC) as a novel ligand at pH 6.5. The complexation between cadmium and ligand was achieved with the MBBOTSC ligand and cadmium extracted by the USA-D-ILLME procedure. By the proposed procedure, the simple and fast extraction, as well as the efficient separation for cadmium ions was obtained at optimized conditions. Results showed the LOD, the working range and RSD ranges were obtained at 0.3 μg L⁻¹, 1-75 μg L⁻¹ and 1.12%-2.54%, respectively. Due to results, the separation cadmium in water samples was simply achieved by the IL phase before determined by AT-F-AAS.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Added</th>
<th>USA-D-ILLME *</th>
<th>Recovery (%)</th>
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<td>Drinking Water</td>
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<td>0.45 ± 0.02</td>
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<td></td>
<td>0.5</td>
<td>0.94 ± 0.05</td>
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<tr>
<td></td>
<td>1.0</td>
<td>1.47 ± 0.07</td>
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</tr>
<tr>
<td>Well water</td>
<td>-----</td>
<td>5.14 ± 0.19</td>
<td>-----</td>
</tr>
<tr>
<td></td>
<td>5.0</td>
<td>9.95± 0.44</td>
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<tr>
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<td>10</td>
<td>15.25± 0.62</td>
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<tr>
<td>Wastewater</td>
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<td>32.31 ± 1.52</td>
<td>98.6</td>
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* Mean of three determinations ± confidence interval (P= 0.95, n=5)
Wastewater prepared from petrochemical industry in Arak and well water from south of Tehran (Share-Ray)
The effect of sample volume for cadmium extraction based on MBBOTSC ligand in water samples with high ions were successfully extracted based on the USA-D-ILLME procedure. The cadmium ions (Cd\(^{2+}\)) was separated and validated by the ET-AAS technique.

The optimum sample volume of 50 mL was selected for cadmium extraction based on the MBBOTSC ligand and cadmium extracted by the USA-D-ILLME procedure. By the proposed methodology, the simple and fast extraction, as well as high recovery for cadmium ions in water results demonstrated the accurate extraction can be validated by ET-AAS (Table 3). The validation of methodology for cadmium extraction and determination in water by spiking samples and wastewater samples was simply achieved by the IL phase separation technique and spiking samples. In this study, the spiked samples of wastewater prepared from petrochemical industry in Arak and well water from south of Tehran (Share-Ray) and wastewater were prepared for this work.

Table 3. The validation of methodology for cadmium extraction and determination in water by spiking samples and comparing to the ET-AAS analyzer

<table>
<thead>
<tr>
<th>Samples</th>
<th>Added ((\mu g) L(^{-1}))</th>
<th>ET-AAS* ((\mu g) L(^{-1}))</th>
<th>USA-D-ILLME* ((\mu g) L(^{-1}))</th>
<th>Recovery (%) ET-AAS</th>
<th>Recovery (%) USA-D-ILLME</th>
</tr>
</thead>
<tbody>
<tr>
<td>Drinking Water</td>
<td>----</td>
<td>0.73 ± 0.03</td>
<td>0.69 ± 0.02</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td></td>
<td>0.5</td>
<td>1.21 ± 0.05</td>
<td>1.22 ± 0.06</td>
<td>96.0</td>
<td>106</td>
</tr>
<tr>
<td>Well water</td>
<td>----</td>
<td>3.76 ± 0.16</td>
<td>3.82 ± 0.18</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td></td>
<td>3.0</td>
<td>6.72 ± 0.29</td>
<td>6.77 ± 0.32</td>
<td>98.6</td>
<td>98.3</td>
</tr>
<tr>
<td>Wastewater</td>
<td>----</td>
<td>14.02 ± 0.71</td>
<td>13.87 ± 0.64</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td></td>
<td>15</td>
<td>28.56 ± 1.28</td>
<td>29.03 ± 1.42</td>
<td>96.9</td>
<td>101.1</td>
</tr>
<tr>
<td>Wastewater</td>
<td>----</td>
<td>16.32 ± 0.78</td>
<td>16.15 ± 0.81</td>
<td>-----</td>
<td>-----</td>
</tr>
<tr>
<td></td>
<td>20</td>
<td>36.25 ± 1.68</td>
<td>35.58 ± 1.74</td>
<td>99.7</td>
<td>97.2</td>
</tr>
</tbody>
</table>

* Mean of three determinations ± confidence interval (\(P=0.95, n=5\))

Wastewater prepared from petrochemical industry in Arak and well water from south of Tehran (Share-Ray)

5. Acknowledgments

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6. References


[25] ACGIH, Documentation of the Threshold Limit Values(TLVs) and Biological Exposure Indices, American Conference of Governmental Industrial Hygienists, 2016 7th Ed. https://www.acgih.org/science/tlv-bei-guidelines/


